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Vol. XI, 1935-36



SIR JAGADIS CHUNDER BOSE

M.A., D.Sc., LL.D., F.R.S., C.S.I., C.I.E.

Born at Mymensing on the 30th November, 1858

We regret to announce the death of
SIR JAGADIS CHUNDER BOSE, Founder and Director
of the Bose Research Institute, and Editor of the
Transactions, on the 23rd November, 1937, at
Giridih.

The proofs of this Volume of the
Transactions were passed by him on the 20th
November, 1937.

TRANSACTIONS of the
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Vol. XI, 1935-1936
BIOLOGICAL AND PHYSICAL
RESEARCHES

EDITED BY
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BIOLOGICAL AND PHYSICAL RESEARCHES

I.—INTRODUCTORY

BY

SIR J. C. BOSE, F.R.S.

A SYNOPSIS of various investigations carried out during the past year in different branches of biophysics, in anthropology and in advanced physics is given below.

Investigation on the effect of age in inducing variation in growth activity of a water plant.—Automatic records of the rate of growth were taken continuously during both night and day for nearly a fortnight, by which time the cycle of growth was completed. The leaf was by this time quite mature, its growth having practically come to a stop.

In the first phase of growth cycle the total growth elongation during 24 hours in a typical specimen was found to undergo a continuous increase till on the ninth day it reached a maximum. After this the rate of growth underwent a continuous diminution till there was total cessation on or about the fifteenth day, when the leaf became fully mature.

In regard to the difference of the rate of growth during night and day, this was found in a typical specimen to be as follows. At the first part of the cycle the rate of growth in daytime was greater than at night. Thus when the leaf was 4 days old the rate of growth during the day was nearly twice

as great as that at night. After this the difference between the two rates became less marked in a progressive manner, till when the leaf was 8 days old the difference in the two rates practically disappeared. Subsequently, when the leaf was 9 days old, the relative rates were found to have undergone a reversal, the rate at night being now the greater of the two. Henceforth the rates both by night and day were found to undergo a continuous diminution till total abolition of growth occurred, when the leaf became fully mature at the age of about a fortnight.

Investigations on the effect of changes of temperature in inducing variation in the rate of growth of a water plant.—Special methods were devised for rapid and accurate determination of variation in the rate of growth under changes of temperature above and below the normal.

Automatic records for this purpose were obtained with the *High Magnification Crescograph*.

It has thus been found that under the environmental conditions to which the plant has been accustomed the rate of growth below the normal temperature undergoes a continuous diminution, till at the critical minimum temperature of about 19°C . there is an arrest of growth.

Under rise of temperature above the normal 30°C . there is an enhancement in the rate of growth, the optimum rate being attained between 33°C . and 36°C . Above this optimum there is a slow diminution in the rate of growth. At 50°C . the plant becomes quite limp and the tissue becomes disintegrated.

*Investigations on the oxygen consumption of the seed of *Cajanus* in the pre-resting, resting and post-resting stages.*—It has been found that the oxygen consumption of the pre-resting seed is continuously decreased along with the decrease of moisture percentage of the seed. The rate of oxygen consumption at about 80 per cent. water content is very high. From about 80 per cent. to 66 per cent. the fall of the rate of oxygen consumption is very rapid, and

after that the rate of fall is progressively lowered till about 12 per cent. of moisture content.

It has also been found that in the resting stage the oxygen consumption is absolutely nil. When the resting stage is passed the post-resting seed begins to absorb oxygen very slowly; the absorption of oxygen, however, increases when the moisture content of the post-resting seed is increased by absorption of water from the atmosphere.

The oxygen consumption of partially soaked post-resting seed is progressively increased along with the increase of water content of the seed. When the fully soaked post-resting seed is gradually brought down to lower moisture contents by dehydration the oxygen consumption is gradually decreased. The consumption of oxygen at this condition is, however, comparatively greater than that of the partially soaked seed of corresponding moisture content.

Investigations on the effects of low atmospheric pressure in modifications of Autonomous Pulsatory Activity and of Moto-Excitability of plants led to the conclusion that the pulsation of *Desmodium* is slightly increased in frequency at a pressure of 560 mm., or 200 mm. below normal. At 460 mm., or 300 mm. below normal, the increase of frequency is more marked. The pulsation is greatly diminished in amplitude and frequency at 360 mm., or 400 mm. below normal, and it became completely arrested when the pressure was reduced to 260 mm., or 500 mm. below normal. The normal pulsatory activity is, however, restored on return to the normal pressure.

Even under reduced pressure which arrests the pulsatory activity the pulsations remain unaffected when the partial pressure of oxygen is maintained under normal atmospheric pressure.

In regard to the moto-excitability of plants, it undergoes a slight decrease at 460 mm., or 300 mm. below

normal. The excitability becomes much diminished at 360 mm., or 400 mm. below normal, and it undergoes an abolition at 260 mm., or 500 mm. below normal pressure. After restoration to the normal pressure there is, however, a complete recovery of its excitability. The excitability also remains unaffected even under a pressure of 260 mm., or 500 mm. below normal, when the partial pressure of oxygen is maintained equal to that of the normal atmosphere.

Chemical examination of the substance obtained from the leaves of Clerodendron infortunatum (Bhant) has been carried out. A non-glucosidal bitter principle has thus been isolated in a chemically pure state. Several derivatives, for example acetyl, benzoyl, phenyl urethane and bromo-compounds, have been prepared and a certain amount of light has been thrown on its chemical constitution. It appears to be an unsaturated hydroxy ester.

A specific colour reaction has been developed by which a minute quantity of *Clerodin* can be detected.

Its anthelmintic property has been verified by physiological experiments *in vitro*.

The seeds of Pachyrhizus angulatus (Sank-alu) have been chemically examined and the different constituents, such as mineral matter, oil content, protein content and carbohydrate content, have been determined. The oil has also been fully analysed. Feeding experiments have shown that the seeds of *Pachyrhizus angulatus (Sank-alu)* are unsuitable as food. They contain a toxic principle which answers to the tests employed for the detection of saponin.

Investigations on the food value of Pisum sativum (Mutur) in the ripened state show that though it can serve as a diet without bringing on any disease in rats, yet the gain in weight of the subjects is generally below normal. Rats when very slightly affected by devitaminised food seem to recover when placed on *Mutur* food. The rats fed on *Mutur* diet are, however, found to be unable to discharge the function of procreation. It would therefore seem that

there is a certain vitamin deficiency in *Mutur* or that there is something in *Mutur* which inhibits or destroys the power of procreation.

Investigations in Genetics have been carried out on the Interspecific Hybridisation in Oryza. The results obtained show that the chromosomes of *O. officinalis* Wall. ($2n = 24$) have been examined and found to be larger in size than the chromosomes of *O. sativa* L. It agrees with *O. sativa* in chromosome number, chiasma frequency, and degree and type of secondary association of the bivalents or univalents at metaphase I or II respectively.

The phenomenon of pollen mother-cell fusion and formation of 'Restitution nucleus' were observed at meiosis in *O. officinalis*. These aberrations lead to diploid gametes formation with 24 chromosomes in each gamete.

The F_1 hybrid of *O. sativa* \times *O. officinalis* shows complete lack of synapsis and presence of univalents. The differences in chromosome size observed in the two parent species are maintained in the hybrid. Shrivelled pollen grains of different sizes and numbers are formed as a result of irregular meiotic divisions.

A detailed study of the somatic chromosomes of *O. minuta* Presl ($2n = 48$) shows that 24 chromosomes are small and 24 chromosomes are larger. These differences in the size of the chromosomes of *O. minuta* are comparable to the differences in size of the chromosomes of *O. sativa* and *O. officinalis*.

O. minuta is characterised by 24 bivalents at meiosis. Secondary pairing occurs among the markedly different sized bivalents or univalents at metaphase I or II respectively. The maximum number of secondary association observed is 12 groups of two bivalents; 12 is therefore the basic number of *O. minuta*.

In the interspecific hybrid *O. minuta* \times *O. officinalis* 12 bivalents are formed which are bigger in size. Pairing is autosyndetic, each of the bivalents being of similar size

and shape. The rest of the 12 chromosomes are smaller in size than the bivalents and are non-homologous.

In the hybrid *O. minuta* \times *O. sativa* 12 bivalents are also formed which are smaller in size than the rest of the 12 univalents. The chromosome pairing is autosyndetic, each bivalent being of similar size and configuration.

A comparison shows that these differences in chromosome size in the two hybrids correspond to the difference in size of the two sets of chromosomes in *O. minuta*.

The chiasma frequency is the same in the pure species and in the hybrids despite the differences in chromosome sizes.

The differences in the size of the chromosomes in the species of *Oryza* are due to genic and structural changes in the morphology of the chromosomes.

The conclusion is drawn that *O. minuta* is an allopolyploid (modified octoploid) originated from a cross between *O. officinalis* and *O. sativa* as a result of diploid gametes formation, but possessing morphologically dissimilar and non-homologous sets of chromosomes.

This theory of the hybrid origin of *O. minuta* is amply confirmed by the following facts :

(a) *O. minuta* is very closely related morphologically to *O. officinalis* and *O. sativa*.

(b) It represents a re-combination of characters from both of these two species.

(c) Geographical distribution of the two species, *O. officinalis* and *O. sativa*, gives ample opportunity for such hybridisation.

(d) Diploid gametes are of frequent occurrence in both the species.

(e) The somatic chromosome number of *O. minuta* Presl ($2n = 48$) has the sum of the somatic chromosome numbers of *O. officinalis* Wall. ($2n = 24$) and *O. sativa* L. ($2n = 24$).

(f) The chromosome pairing is autosyndetic in meiosis, as a result of which *O. minuta* is fully fertile and breeds true to type.

The Anthropological investigations have been carried out on the racial characteristics of the Rajmahal aborigines. The results obtained show that the only difference in the three groups of the aborigines lies in the stature, which is most probably due to their changes in habitats. The difference between the Hill Mālér's and the Plains Mālér's is considerable in the Transverse arc and Transverse Cephalofacial index of the head ; slight differences occur, however, in Auricular height, in Bigonial breadth, and in Inter-Orbital breadth. The difference between the Hill Mālér's and the Mālpāhāriās is great in Auricular height, in Orbitonasal breadth, in Upper facial length, in Horizontal Circumference of head, in Transverse arc of head, in Nasal index and in Vertical Cephalofacial index. There are, however, slight differences in Bigonial breadth, in Total facial length, in Sagittal arc of the head, Nasal Elevation index and in Transverse Cephalofacial index. The above shows that the Mālpāhāriās differ more from the Hill Mālér's than the latter differ from the Plains Mālér's. It would appear that the Mālpāhāriās must have separated a very long time ago and that the changes due to the environments of the plains have considerably influenced them.

In regard to Mālér-Oraon affinities it is found that appreciable differences exist between them in regard to Stature, Auricular height of head, Orbitonasal breadth, Orbitonasal arc, Total facial length, Sagittal arc of head, Transverse arc of head, Length-height index, Breadth-height index, Nasal Elevation index, Total facial index and Vertical Cephalofacial index. That there is also some difference in the form of the head is evident from the large difference in the head-height and in the length of the head ; this latter is as many as six times the difference between the

Hill Mālér's and the Plains Mālér's, and ten times that between the Hill Mālér's and the Mālpāhāriās. It has already been shown in what ways the Oraons differ characteristically from the Mālér's, and it would therefore be difficult to assume any close relationship between the two. The dissimilarity of the cultures of the two people also offers an additional proof of the difference between them. It would thus appear that the Mālér's and the Oraons are two independent Dravidian-speaking peoples who never came into intimate contact with one another. The Mālér's may be taken as the autochthonous inhabitants of these hills, where they are still found to be living; they undoubtedly appear to be one of the earliest remnants of the pre-Dravidian tribes inhabiting this particular region.

In the Department of Physics investigations were carried out in the following subjects:

On the absorption band in aqueous solution of alkali halide, indicating that the particular band seems to be due to electro-affinity spectrum of water.

On the variation in the intensity of Cosmic Rays and their distribution at the different altitudes of Calcutta and Darjeeling, as also from different azimuthal and of different zenith angles. Comparison has been made with results obtained by other workers in other parts of the earth.

The problem of the propagation of Radio Waves in the upper ionised region of the earth's atmosphere has been subjected to a rigorous and quantitative mathematical investigation based on the new quantum mechanics.

II.—VARIATION IN LONGITUDINAL GROWTH OF A WATER PLANT DURING ITS LIFE CYCLE

BY

S. C. DAS, M.A., AND B. K. PALIT, B.Sc.

THE study of variation in growth during the life cycle of land plants has attracted the attention of many workers. It has been observed that the growth of land plants during their life cycle, instead of being a continuous process of uniform elongation, exhibits a series of wave-like variations. In our recent work on the 'Periodic Variation in Longitudinal and Diametric Growth of Stem in *Helianthus*'¹ it was shown that the longitudinal and diametric growths of the plant during its life exhibited periodic variations consisting of series of distinct maxima.

The nature of variation of growth of a completely submerged water plant during its entire life cycle is, however, very little known. An accurate and quantitative determination of the changes in its growth is a very difficult problem. For the physical conditions relating to the environment under which water plants grow differ considerably from those of land plants in regard to both thermal condition and to intensity of illumination.

The present investigation on a water plant was undertaken in order to obtain better knowledge in regard to the variation of the rate of the longitudinal growth of the leaf during the different periods of its life cycle. For this purpose the water plant *Vallisneria spiralis* was selected

¹ S. C. Das and B. K. Palit, 'Periodic Variation in Longitudinal and Diametric Growth of Stem in *Helianthus*,' *Trans. Bose Research Institute*, vol. ix (1933-34), pp. 35-57.

for the experiment, since the life cycle of an individual leaf of this plant is completed during as short a period as a fortnight; the effect of any change which might result from the variation of season is thus avoided.

CHARACTERISTICS OF THE WATER PLANT

Vallisneria spiralis grows completely submerged in water. The plant has a short stem which is about 2 inches in length. The leaf is ribbon-like, and when moderately grown is about $\frac{1}{4}$ inch in breadth and $1\frac{1}{2}$ to 2 feet in length. It has no nodes and internodes like the submerged water plant *Hydrilla*. It is mainly reproduced in a vegetative manner through numerous stolons.

EXPERIMENTAL ARRANGEMENTS

The plant being completely submerged in water, the method employed for recording the changes in its longitudinal growth at different periods of its life necessitated special contrivances. Specimens that were grown in the special tanks of the Institute were taken out with intact roots and adhering mud. They were then transferred to glass beakers about 4 inches in height and 3 inches in diameter. In order that the plant might be kept undisturbed at the bottom of the vessel the stem of the plant was attached to a heavy base. For this purpose a conical vessel of glass about 2 inches in height and filled with lead shot was employed. The upper end of this glass vessel was drawn into the form of a cylinder terminating laterally in two hooks, and then sealed. The stem of the rooted plant was fastened securely to the cylindrical neck of the vessel by means of a silk thread. The plant thus mounted in a beaker was placed under water in a rectangular glass vessel, the height of which was 24 inches, the length and breadth being 12 inches and 8 inches respectively. The level of water in the outer rectangular vessel of glass was, to start with, adjusted to be $1\frac{1}{2}$ inches above the tip of the experimental

leaf blade. As the leaf grew upwards fresh water was added once in every 24 hours, so as to maintain the water level above the tip of the leaf approximately constant.

The experiments were carried out and records obtained in a glass house provided with suitable arrangements for free ventilation. The diurnal record of growth was obtained by means of the *Oscillating Recorder*. The vessel containing the plant and the Recorder was further enclosed in a glass case to prevent disturbance which might be caused by currents of air. The top of this glass case was covered over with brown paper to prevent direct rays of the sun falling on the plant.

A diagrammatic representation of the complete apparatus together with the plant vessel is given in fig. 1. A specially light aluminium clamp was attached to the tip of the leaf. The clamp was shaped in the form of a triangle provided with a hole below the upper angular notch. The specimen was next attached to the writing lever by glass links of appropriate length, the upper end of the link resting on the S-shaped hook suspended by waxed silk thread from the lever. The point of the writer was brought to the desired position on the recording plate by the adjustment of the fork on which the axis of the lever is pivoted. Dots were automatically inscribed on the recording plate kept oscillating by means of clockwork, the successive dots being at intervals of 12 minutes. The plate moved laterally through 6 inches in 24 hours. The magnification employed in the experiments of this series was only $2\frac{1}{2}$ times, since during the phase of maximum growth a higher magnification would have carried the record outside the plate.

The temperature in the glass house as well as in the plant vessel containing water was simultaneously observed with the help of two maximum and minimum thermometers. The diurnal variation of temperature in the glass house was found to vary through about 8°C. , whereas in the plant vessel itself the variation was only about 4°C.

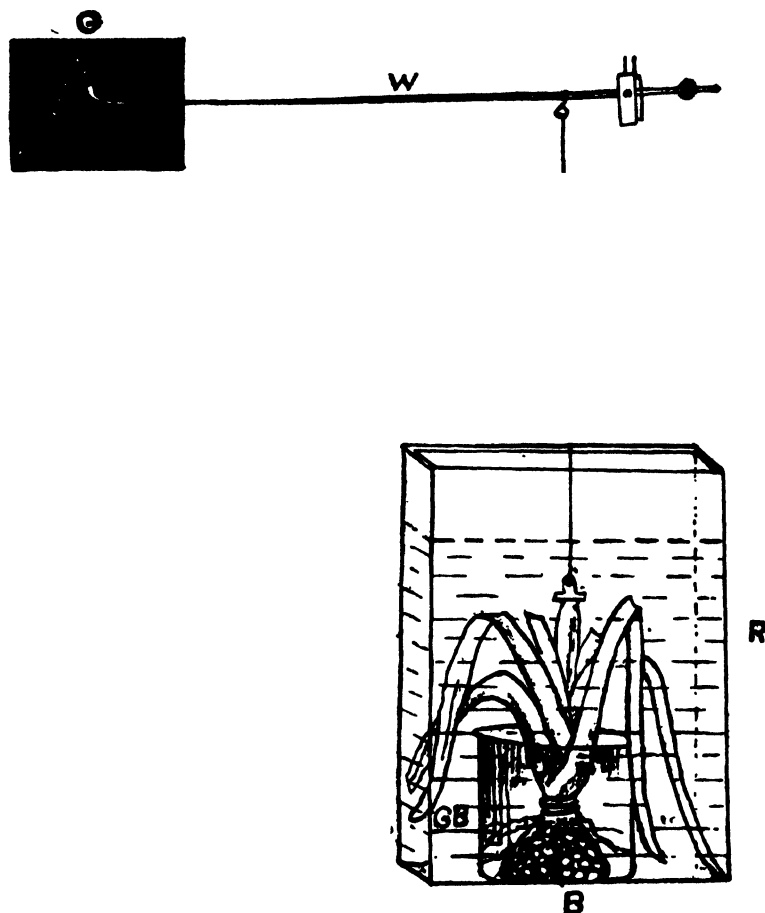


FIG. 1. Semi-diagrammatic representation of the complete apparatus for determination of growth variation of *Vallisneria spiralis* throughout the life cycle.

The entire plant with roots intact securely fastened to a heavy base (B) is placed in a glass beaker GB. The plant thus mounted is placed in the rectangular water trough R. The tip of the plant is appropriately connected by suitable glass links with a S-shaped hook suspended from the writing lever W. Growth is recorded on the smoked-glass plate G as an up-curve.

Successive dots in this and in the following records are at intervals of 12 minutes.

Magnification employed is $2\frac{1}{2}$ times.

Experiments were performed with six different specimens of *Vallisneria spiralis*, records being taken of the diurnal growth elongation throughout their entire life cycle. Typical results secured from the experiments will be given presently.

The age of the particular specimen was calculated from the time which elapsed between the date of appearance of the leaf on the axis of the stem and the day of experiment.

Determination of longitudinal growth was made every day, commencing on September 17 and continuing up to September 29, 1936. In this way 12 records were obtained with an identical specimen for determination of growth variation during the whole cycle, which, as previously stated, was completed in about a fortnight. The length of the specimen when 4 days old was 18.75 mm.

Experiment 1. *Determination of longitudinal growth of the leaf of Vallisneria spiralis when 4 days old.*—The first record of the series was taken from 6 P.M. on September 17 to 6 P.M. on September 18. The total increase of length in 24 hours is found from the magnified record to be 65 mm. (fig. 2). As the magnification employed was $2\frac{1}{2}$ times, the actual growth in length in 24 hours is therefore 26 mm.

As regards the relative rate of growth during the night and during the day, this can be deduced from the curve itself, in which the first half represents growth elongation during the night from 6 P.M. to 6 A.M. The second half of the curve represents growth during the daytime between 6 A.M. and 6 P.M. As the slope of the first part of the curve (representing rate of growth at night) is relatively slight, it is clear that the rate of growth at night is slower than that during the day. For obtaining quantitative results it is to be remembered that the recording lever magnifies $2\frac{1}{2}$ times. The total magnified growth at night is 22.5 mm., the actual growth elongation being 17 mm. Hence the average rate of growth during the day is nearly twice as great as that at night.

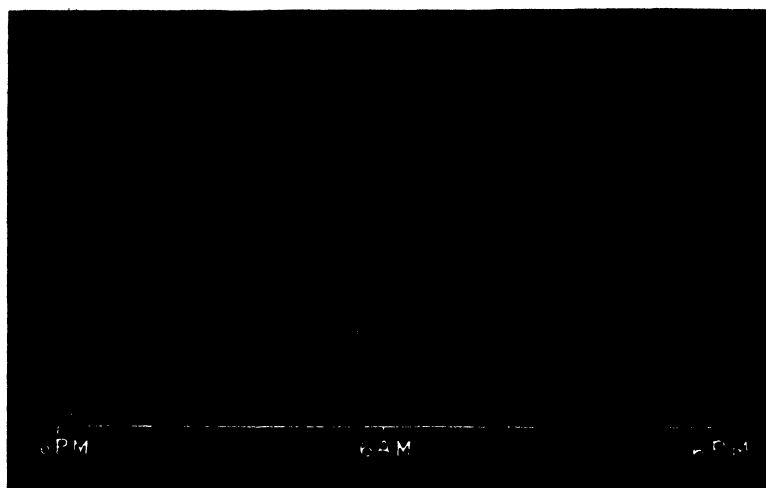


FIG. 2. Record of longitudinal growth of *Vallisneria spiralis* when 4 days old.

The abscissa represents the hours of day and night, the ordinate representing growth elongation. The growth at any hour is found from the vertical distance of the curve from the point in the base representing the hour.

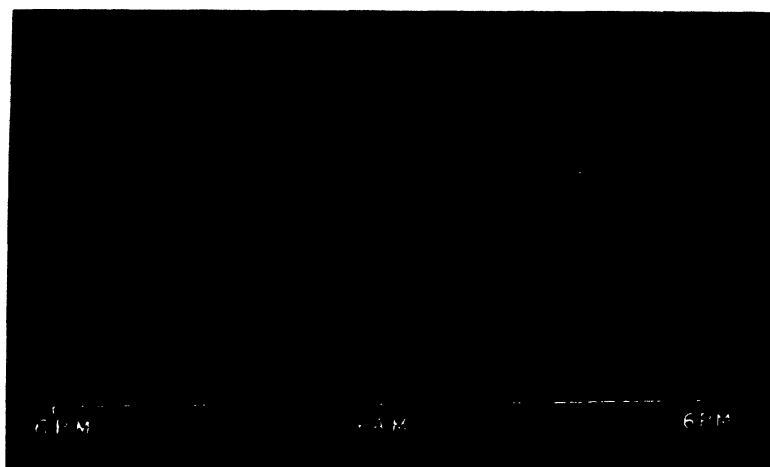


FIG. 3. Record of longitudinal growth of *Vallisneria spiralis* when 5 days old.

Experiment 2. *Longitudinal growth of the leaf of Vallisneria spiralis 5 days old.*—In fig. 3 is given the next record of growth obtained during the following 24 hours from 6 P.M. on September 18 to 6 P.M. on September 19. Following the method of calculation already described, it is found that the actual growth elongation in 24 hours is 26 mm.

As regards the relative rates of growth during night and day, the average rate during the night is 10.5 mm., while

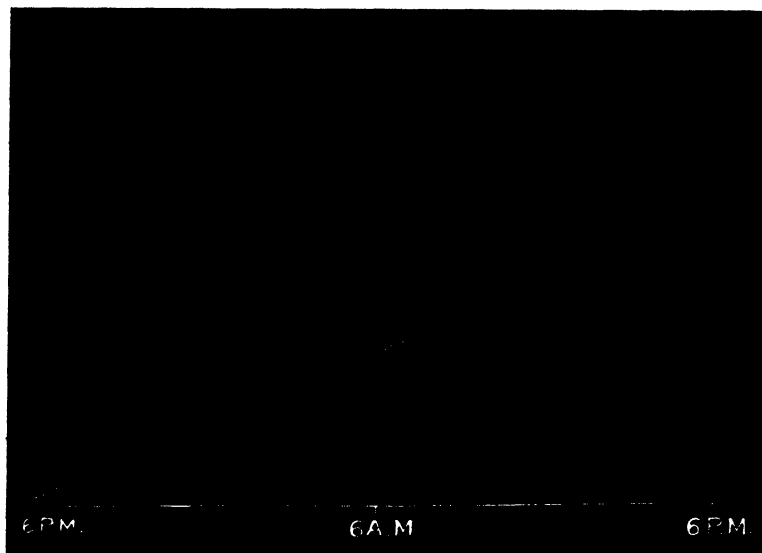


FIG. 4. Record of longitudinal growth of *Vallisneria spiralis* when 6 days old.

that during the day is 15.5 mm. The rate of growth during the day is therefore 1.5 times as great as that during the night.

Experiment 3. *Longitudinal growth of the leaf of Vallisneria spiralis 6 days old.*—The third record of the series (fig. 4) was taken between September 19 and September 20. The total longitudinal growth in 24 hours is found to be 32 mm., which is 6 mm. greater than that on the previous day.

As regards the relative growth elongations during the night and during the day, the growth elongation at night is 12.75 mm., while that during the day is 19.25 mm. The average rate of growth during the day is therefore 1.5 times greater than that during the night.

Experiment 4. *Longitudinal growth of the leaf of Vallisneria spiralis 7 days old.*—In fig. 5 is shown the record

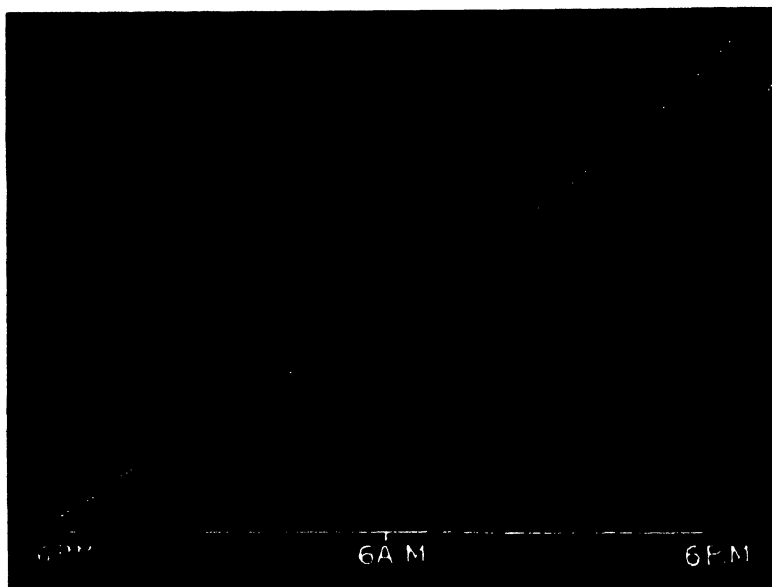


FIG. 5. Record of longitudinal growth of *Vallisneria spiralis* when 7 days old.

of growth obtained between September 20 and September 21. The more erect slope of the curve indicates that during the first days of the cycle there is an increase in the rate of growth with the age of the specimen. The total growth elongation during 24 hours is 43 mm., which is considerably greater than that on the previous days. Again, the growth elongation during the night is 19 mm., while during the day it is 24 mm., or 1.3 times greater than that during the night. The difference

between the rates of growth during night and day is thus becoming less pronounced.

Experiment 5. *Longitudinal growth of the leaf of Vallisneria spiralis 8 days old.*—The next record of the series, taken between September 21 and September 22, is represented in fig. 6. The actual increase of length as calculated

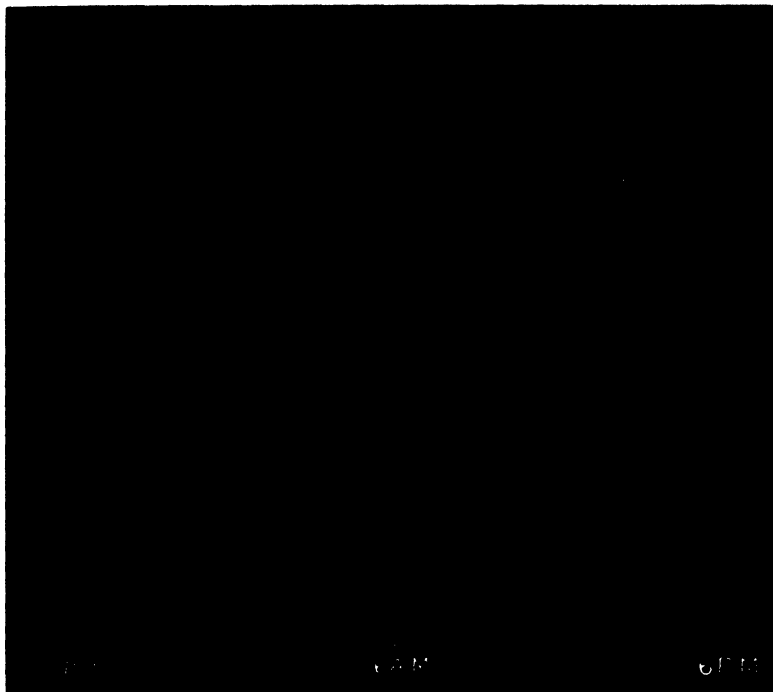


FIG. 6. Record of longitudinal growth of *Vallisneria spiralis* when 8 days old.

from the record is 49 mm. The growth activity is thus seen to be steadily increasing with the age of the leaf. The respective growth elongations during night and day are found to be 24 mm. and 25 mm. respectively. Thus at a certain age of the leaf the difference between the night and day growth rates is tending to disappear.

Experiment 6. *Longitudinal growth of the leaf of Vallisneria spiralis 9 days old.*—In fig. 7 is given the record of growth obtained between September 22 and September 23. The growth activity is seen to be further enhanced with increase of the age of the leaf, the actual growth elongation during 24 hours being 53 mm. The growth on the 9th day

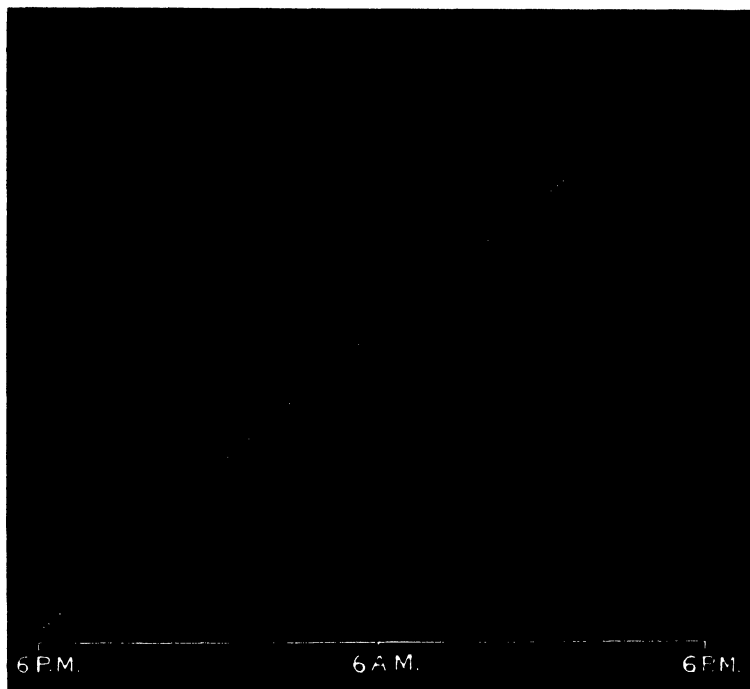


FIG. 7. Record of longitudinal growth of *Vallisneria spiralis* when 9 days old.

will presently be found to have reached the maximum. Again, in regard to the relative growths during the night and the day, the total growth elongation during the night is 28 mm., while that during the day is only 25 mm. On the 9th day, therefore, the rate during the night, instead of being smaller, is actually greater than that during the day.

Experiment 7. *Longitudinal growth of the leaf of Vallisneria spiralis 10 days old.*—The next record of the series, obtained between September 23 and September 24, is represented in fig. 8. The growth activity, after attaining a maximum on the 9th day, is seen to undergo a diminution with further increase of age. The actual increment in

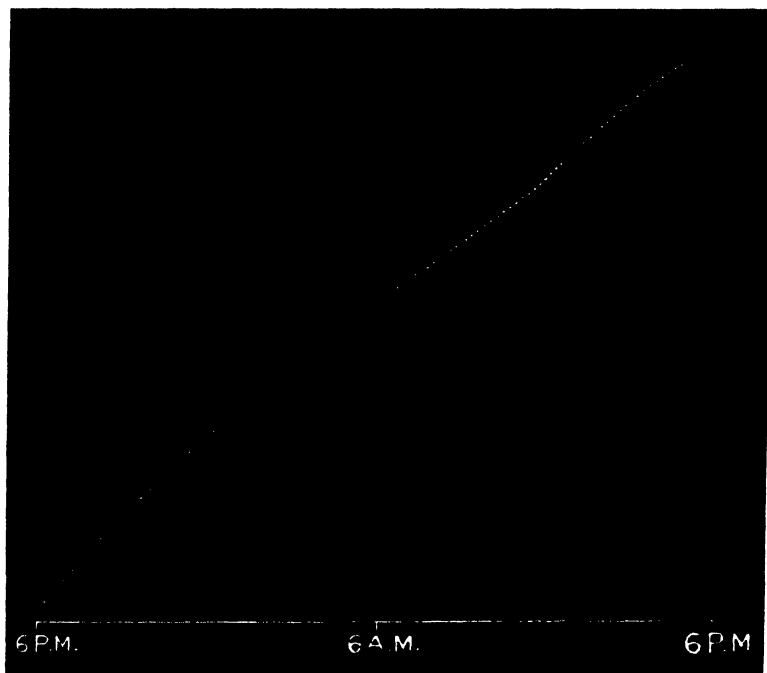


FIG. 8. Record of longitudinal growth of *Vallisneria spiralis* when 10 days old.

length during 24 hours on the 10th day, as calculated from the record, is 49 mm., which is 4 mm. less than that on the previous day.

As the slope of the first part of the curve (representing rate of growth at night) is relatively steeper, it is clear that the rate of growth at night is greater than that during the day. The growth elongations during night and day are found to be 27.5 mm. and 21.5 mm. respectively.

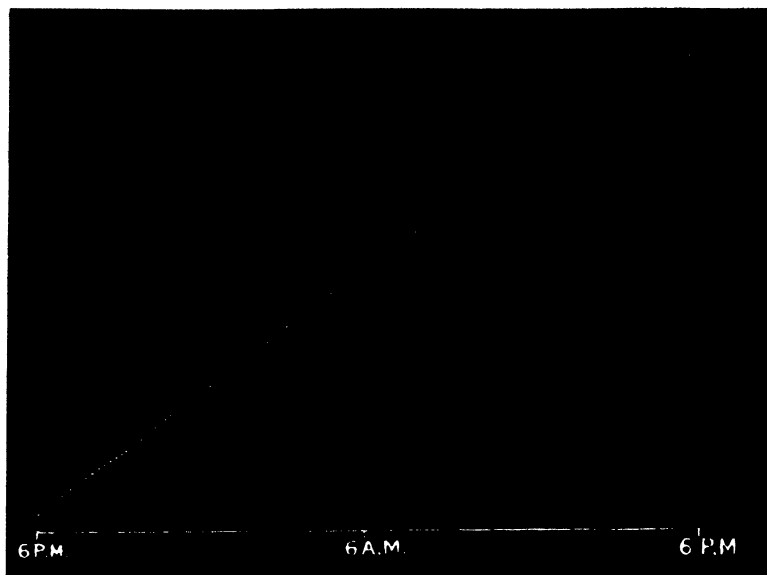


FIG. 9. Record of longitudinal growth of *Vallisneria spiralis* when 11 days old.

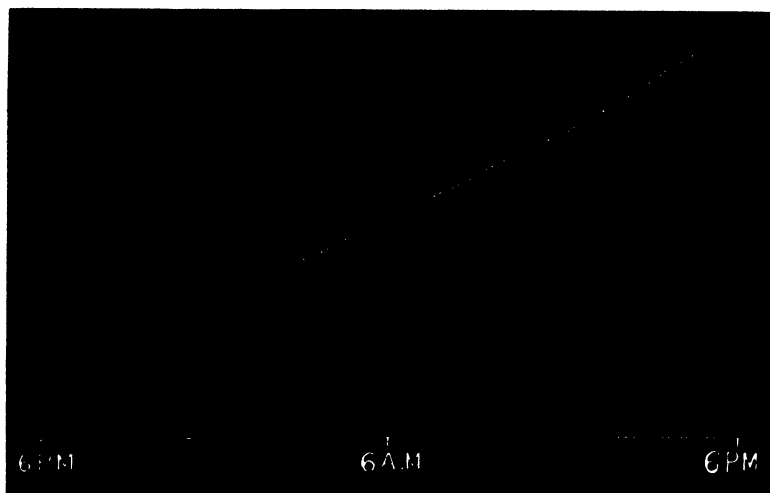


FIG. 10. Record of longitudinal growth of *Vallisneria spiralis* when 12 days old.

Experiment 8. *Longitudinal growth of the leaf of Vallisneria spiralis 11 days old.*—In fig. 9 is shown the record of growth obtained between September 24 and September 25. The growth activity is seen to be further decreased with the age of the leaf, the actual growth elongation during 24 hours on the 11th day being 40 mm., which is 9 mm. less than that on the previous day. The respective rates of growth during the night and during the day are found to be 21.5 mm. and 18.5 mm. Here also the rate of growth at night is greater than that during the day.

Experiment 9. *Longitudinal growth of the leaf of Vallisneria spiralis 12 days old.*—In fig. 10 is given the record of growth obtained between September 25 and 26. The growth activity is seen to be further lowered with the increase of the age of the leaf, the actual growth elongation during 24 hours on the 12th day being 30 mm. The relative rates of growth during the night and the day are 16 mm. and 14 mm. respectively. The difference between the rates of growth during night and day is thus becoming less pronounced.

Experiment 10. *Longitudinal growth of the leaf of Vallisneria spiralis 13 days old.*—The next record of growth obtained between September 26 and 27 is represented in fig. 11. The activity is seen to be further diminished with the increase of the age of the leaf, the actual increment in length during 24 hours on the 13th day being 19 mm. In regard to the relative growths during the night and the day, the total growth during the night is 10.5 mm., while that during the day is only 8.5 mm.

Experiment 11. *Longitudinal growth of the leaf of Vallisneria spiralis 14 days old.*—In fig. 12 is shown the record of growth obtained between September 27 and 28. The growth activity is found to have undergone a further diminution with the increase of the age of the leaf, the actual growth elongation during 24 hours on the 14th day being 10 mm. The night and day growth elongations

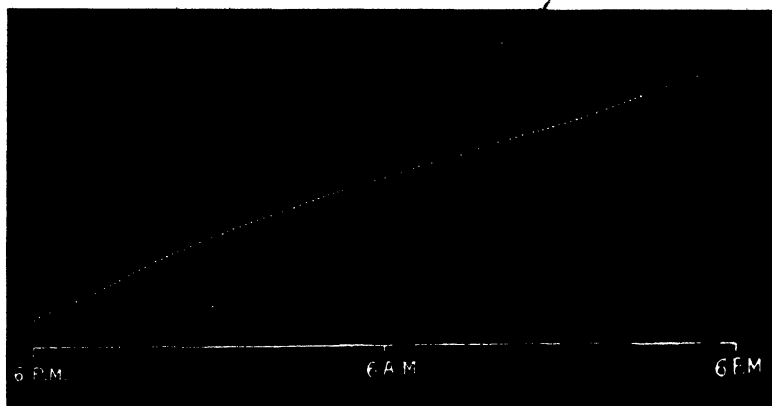


FIG. 11. Record of longitudinal growth of *Vallisneria spiralis* when 13 days old.

FIG. 12. Record of longitudinal growth of *Vallisneria spiralis* when 14 days old.

FIG. 13. Record of longitudinal growth of *Vallisneria spiralis* when 15 days old.

are thus found to be 5.5 mm. and 4.5 mm. respectively. It will be seen that when the leaf approaches maturity the difference between the rates of growth during night and day tends to disappear.

Experiment 12. *Longitudinal growth of the leaf of Vallisneria spiralis 15 days old.*—The last record of the series, given in fig. 13, was taken between September 28 and 29. The growth activity of this mature leaf is seen to be almost abolished, the actual increment in length during 24 hours on the 15th day being 2 mm. The relative rates of growth during the night and the day are 2 mm. and 0 mm. respectively.

The summary of results of variation in longitudinal growth with the age of a specimen of *Vallisneria spiralis* is seen detailed in the following table.

TABLE I.—DIURNAL VARIATION IN LONGITUDINAL GROWTH DURING THE LIFE CYCLE OF A SPECIMEN OF *VALLISNERIA SPIRALIS*. (SPECIMEN I.)

Days of observation	Longitudinal growth in mm., 6 P.M. to 6 A.M.	Longitudinal growth in mm., 6 A.M. to 6 P.M.	Total elongation in 24 hours in mm.	Average rate of 24 hours in μ per second
1	9.0	17.0	26.0	0.30
2	10.5	15.5	26.0	0.30
3	12.75	19.25	32.0	0.37
4	19.0	24.0	43.0	0.50
5	24.0	25.0	49.0	0.57
6	28.0	25.0	53.0	0.61
7	27.5	21.5	49.0	0.57
8	21.5	18.5	40.0	0.46
9	16.0	14.0	30.0	0.34
10	10.5	8.5	19.0	0.22
11	5.5	4.5	10.0	0.12
12	2.0	0.0	2.0	0.023
Mean .	15.5	16.0	31.5	0.36

In the table on p. 23 the first column gives the day of observation, the first of which was taken when the leaf was 4 days old. The fourth column gives the changes in the rate of growth with the increasing age of the specimen. On the first day of observation of the leaf, 4 days old, the total growth elongation was 26 mm. When the leaf was 5 days old, the total growth remained the same. On the third day of observation, when the leaf was 6 days old, the total growth elongation had undergone a considerable increase, being 32 mm. The growth rate subsequently underwent a continuous increase till the maximum rate was attained when the leaf was 9 days old, the total growth elongation being now 53 mm. After this, further increase of age is followed by continued diminution of the rate of growth, which was practically abolished when the leaf was fully mature on the 15th day.

The curve (fig. 14 below) gives a graphic representation of the relation between the diurnal longitudinal growth and the age of the leaf.

In regard to the difference between the rates of growth during night and the day, the rate in the typical specimen (1) is at first greater during the day. When the leaf was 4 days old, the rate during the day was nearly twice as great. On the next day the difference was less, and the diminution of the difference went on progressively till, when the leaf was 8 days old, the difference in the rates practically disappeared. On the next day, when the leaf was 9 days old, the relative rates underwent a reversal, the rate at night being now greater. After this rates of growth both at night and day underwent a continuous diminution till, on attaining the age of about 15 days, growth came practically to a stop, the leaf being fully matured.

The results obtained with two other specimens are given in Tables II and III. These may be regarded on the whole as confirming the results obtained from Specimen 1.

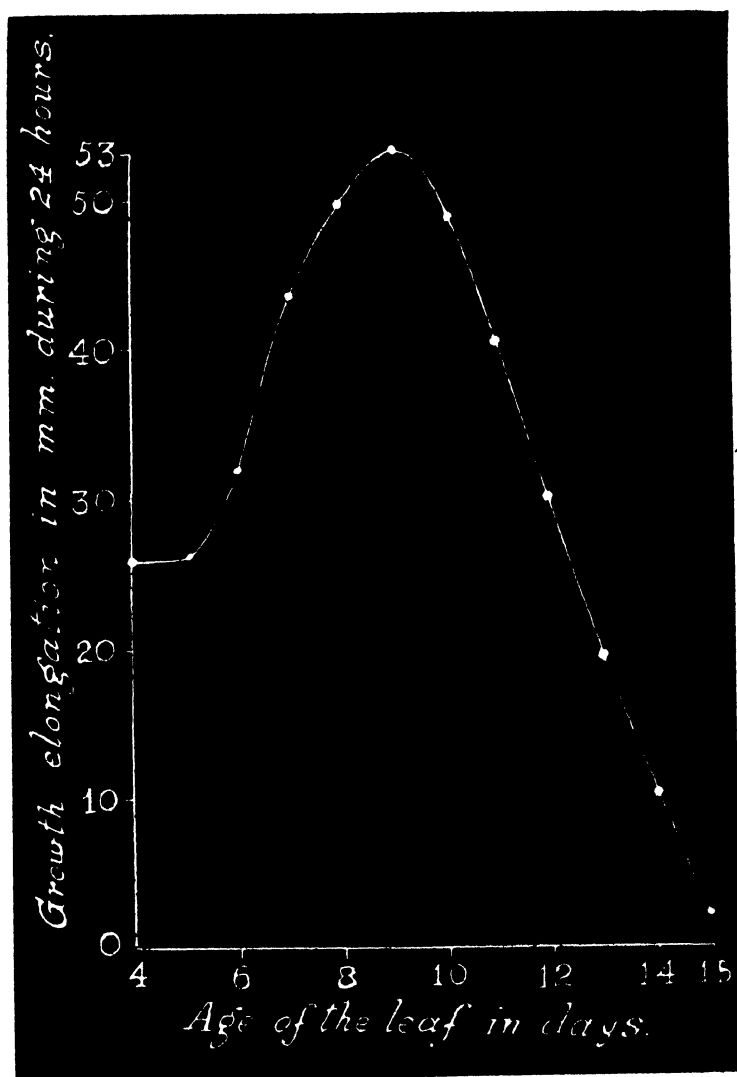


FIG. 14. Graphic representation of the relation between the diurnal longitudinal growth and the age of the leaf.

TABLE II.—DIURNAL VARIATION IN LONGITUDINAL GROWTH DURING THE LIFE CYCLE OF *VALLISNERIA SPIRALIS*. (SPECIMEN 2.)

Days of observation	Longitudinal growth in mm., 6 P.M. to 6 A.M.	Longitudinal growth in mm., 6 A.M. to 6 P.M.	Total elongation in 24 hours in mm.	Average rate of 24 hours in μ per second
I	5.0	8.0	13.0	0.15
2	6.5	9.5	16.0	0.18
3	7.5	12.5	20.0	0.23
4	13.5	16.5	30.0	0.34
5	19.5	22.5	42.0	0.48
6	25.5	23.5	49.0	0.57
7	24.5	18.5	43.0	0.50
8	19.5	16.5	36.0	0.41
9	15.0	12.0	27.0	0.31
10	9.0	8.0	17.0	0.20
11	4.0	3.5	7.5	0.08
12	2.0	1.0	3.0	0.034
Mean .	12.6	12.6	25.3	0.29

TABLE III.—DIURNAL VARIATION IN LONGITUDINAL GROWTH DURING THE LIFE CYCLE OF *VALLISNERIA SPIRALIS*. (SPECIMEN 3.)

Days of observation	Longitudinal growth in mm., 6 P.M. to 6 A.M.	Longitudinal growth in mm., 6 A.M. to 6 P.M.	Total elongation in 24 hours in mm.	Average rate of 24 hours in μ per second
I	9.0	12.0	21.0	0.24
2	9.5	13.0	22.5	0.26
3	14.0	16.0	30.0	0.34
4	17.0	16.0	33.0	0.38
5	20.0	15.0	35.0	0.40
6	16.5	15.5	32.0	0.37
7	13.5	12.5	26.0	0.30
8	10.5	8.5	19.0	0.22
9	8.0	6.0	14.0	0.16
10	5.0	3.0	8.0	0.09
11	1.0	0.5	1.5	0.017
Mean .	11.2	10.7	22.0	0.25

SUMMARY

Investigation on the effect of age in inducing variation in growth activity of a water plant was undertaken with the leaf of *Vallisneria spiralis*.

Automatic records of the rate of growth were taken continuously during both night and day for nearly a fortnight, by which time the cycle of growth was completed. The leaf was by this time quite mature, its growth having practically come to a stop.

In the first phase of growth cycle the total growth elongation during 24 hours in a typical specimen was found to undergo a continuous increase, till on the 9th day it reached a maximum. After this the rate of growth underwent a continuous diminution till there was total cessation on or about the 15th day, when the leaf became fully mature.

The difference in the rates of growth during night and day was found in a typical specimen to be as follows. In the first part of the cycle the rate of growth in the daytime was greater than at night ; when the leaf was 4 days old the rate of growth during the day was nearly twice as great as that at night. After this the difference between the two rates became less marked in a progressive manner, till when the leaf was 8 days old the difference in the two rates practically disappeared. On the next day, when the leaf was 9 days old, the relative rates were found to have undergone a reversal, the rate at night being now the greater of the two. Henceforth the rates both at night and day were found to undergo a continuous diminution, till total abolition of growth occurred when the leaf, at the age of about a fortnight, became fully mature.

We take this opportunity of expressing our grateful thanks to Sir J. C. Bose for his kind suggestions and encouragement, which have been extended to us throughout this investigation.

III.—EFFECT OF VARIATION OF TEMPERATURE ON GROWTH OF A WATER PLANT

BY

S. C. DAS, M.A., AND B. K. PALIT, B.Sc.

IN his work on 'Growth and Tropic Movements of Plants' Sir J. C. Bose described various contrivances by which the effect of variation of temperature on the rate of longitudinal growth of land plants could be quickly determined with the highest degree of accuracy.¹ It was found that the rate of growth increased with the rise of temperature up to an optimum, above which the rate underwent a depression. At lower temperatures below the normal, moreover, the rate was found to decrease, culminating in an arrest at a critical minimum temperature. The question now arises whether a submerged water plant would behave in an analogous manner. If so, would it be possible to obtain accurate and instantaneous records from which the characteristic variations induced under changes of temperature in a submerged water plant could be deduced? The difficulties in solving the problem were numerous and highly complicated. The difficulties have, however, been completely overcome by the construction of a device by which the variation of growth induced by changes of temperature can be automatically recorded without any loss of time.

The records were obtained in a closed room where light from outside could not enter. Only a feeble and constant illumination was maintained by means of a small electric

¹ Sir J. C. Bose, *Growth and Tropic Movements of Plants* (1929), pp. 36-37.

bulb. Means were also taken so that different temperatures to which the plant was subjected could be maintained constant during the period of a particular experiment. The experiment was carried out with the leaf of *Vallisneria*. The short stem of the plant was suitably mounted on a glass stand and all the leaves were cut off except the one to be experimented upon.

METHOD FOR PRODUCING VARIATION OF TEMPERATURE

For this purpose the plant was placed in a light aluminium vessel, V, filled with water. This inner vessel was next placed in a cylindrical glass vessel filled with water and provided with two side tubes, one at the top, t_2 , and the other at the bottom, t_1 , serving as outlet and inlet pipes. There is also a reservoir, R, placed at a higher level, the reservoir containing either hot or cold water, according to different requirements. The temperature of the water in the outer cylindrical vessel can be gradually raised or lowered by regulating the flow of hot or cold water from the reservoir by proper manipulation of stop-cock S. There is a flow of heat from the outer to the inner vessel across the conducting aluminium wall.

The temperature of the inner aluminium cylinder containing the specimen can thus be gradually raised, lowered or maintained at a given temperature. When variation of temperature was found necessary, the rate of change was only 1°C . per minute, for a quicker rate might have induced an excitatory reaction vitiating the true effect.

HIGH MAGNIFICATION CRESCOGRAPH

One of the difficulties of the investigation lay in the extraordinarily slow rate of growth of the leaf, necessitating a prolonged period of experimentation for detection of normal growth and its induced variation, during which time

it was impossible to maintain external conditions constant. This difficulty has been overcome by the invention of the

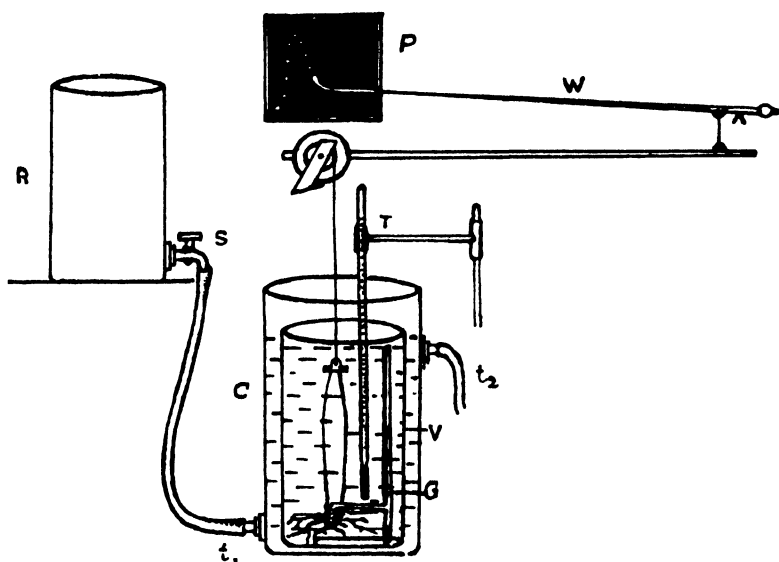


FIG 15 Semi-diagrammatic representation of the complete apparatus for determination of variation of rate of growth of *Vallisneria spiralis* under different temperatures

The plant with roots intact is attached to a glass stand G, and all leaves except one cut off. The specimen is placed in a metallic vessel V in which a thermometer T is inserted. The plant thus mounted in the metallic vessel V is placed within an outer cylindrical glass vessel C, with two side tubes t_1 and t_2 . The tube t_2 is connected with the reservoir R, containing either hot or cold water. The tube of the reservoir is provided with a stop-cock S. The tip of the experimental leaf is connected by a suitable glass link with a high magnification system of compound levers diagrammatically represented in the figure.

Successive dots in this and in the following records are at intervals of 15 seconds.

Magnification employed is 2,500 times.

High Magnification Crescograph.¹ The tip of the leaf was suitably attached to a system of compound levers, a diagrammatic representation of which is given in fig. 15.

¹ Sir J. C. Bose, *Growth and Tropic Movements of Plants*, p. 11.

In the present case the magnification employed was 2,500 times, and the record of magnified growth was taken on an oscillating plate moving to and fro at an interval of 15 seconds. The smoked-glass plate was kept in a particular position and a dotted tracing was obtained, growth being recorded as an up-curve. After the completion of the first series of records the plate was pushed laterally through a short distance, when a second series was obtained. In all

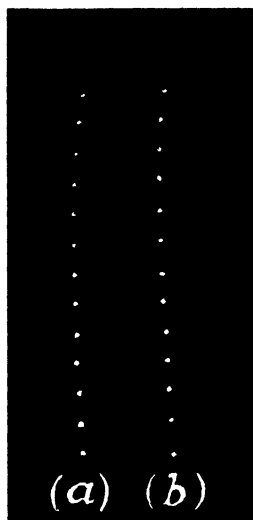


FIG. 16. Recorded growth of *Vallisneria spiralis*. The series (b) was obtained an hour after the series (a).

experiments the specimens chosen were of short length of about 25 to 27 mm.

In order to demonstrate that during the short period of a given experiment the rate of growth of the leaf of *Vallisneria* remained constant under uniform external conditions the following experiment was carried out.

Experiment 1. *Demonstration of constant rate of growth of the leaf under uniform external conditions.*—The temperature of the experimental room was 30° C. and the leaf was

illuminated by a feeble constant light. The external factors were maintained constant for the period of one hour, which is the longest period necessary to conclude any series of experiments. In the present case the first record (fig. 16 (a)) was taken at the beginning and the second (fig. 16 (b)) at the end of the hour. In each of the records (a) and (b) there are 12 intervals between successive dots, each interval representing 15 seconds. The total duration of record is therefore 12×15 or 180 seconds = 3 minutes. The total magnified growth elongation in the two records, the second taken after the interval of an hour, is thus seen to be equal.

Hence it may be concluded that the rate of growth remains constant for at least an hour.

From the records thus secured it is possible to determine the absolute rate of growth under a particular condition. For this we adopt a second as the unit of time and μ or micron as the unit of length, the micron being 0.001 mm.

If m be the magnifying power of the compound lever, and l the average distance between successive dots at intervals of t seconds, then :

$$\text{rate of growth} = \frac{l}{m \times t} \times 10^3 \mu$$

Referring to the record given in fig. 16 :

The magnified growth elongation for 12 dot intervals = 45 mm.

l or average distance between successive dots

$$= \frac{45}{12} \text{ mm.} = 3.75 \times 10^3 \mu$$

$t = 15$ seconds.

$m = 2500$.

$$\text{Absolute rate of growth} = \frac{3.75 \times 10^3 \mu}{2500 \times 15} = 0.1 \mu \text{ per sec.}$$

Experiment 2. Effect of higher temperatures.—The experiment was carried out with a leaf of *Vallisneria*. Hot water from the reservoir was allowed to flow into the outer

glass cylinder and the rise of temperature was adjusted successively to different temperatures. These different temperatures were maintained constant for about five minutes to secure these different records. Records obtained at temperatures of 30°C. , 33°C. , 36°C. , 40°C. and 45°C.

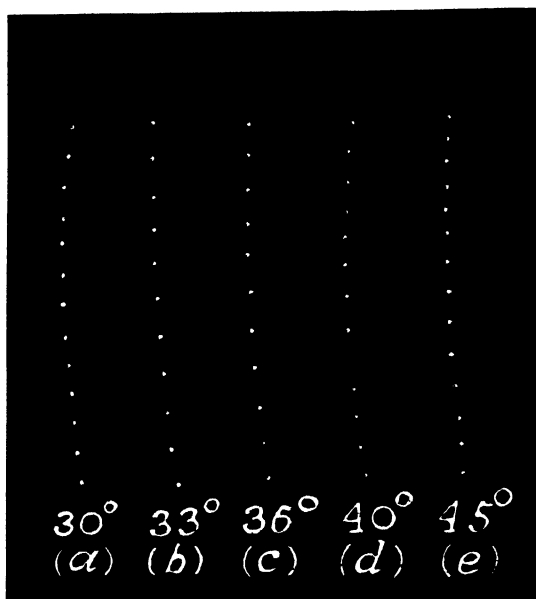


FIG. 17. Effect of higher temperatures on growth of the leaf of *Vallisneria spiralis*.

- (a) Record of normal rate.
- (b) Record of effect of temperature at 33°C.
- (c) Record of effect of temperature at 36°C.
- (d) Record of effect of temperature at 40°C.
- (e) Record of effect of temperature at 45°C.

are reproduced in fig. 17; the time interval between dots is the same; but for the completion of the same length of growth elongation the number of dot intervals is different. At 30°C. the number of dot intervals is 12, at 33°C. it is 10, at 36°C. it is 10, at 40°C. it is 12, and at 45°C. it is 13. The records (fig. 17) show that the rate of growth increases with the rise of temperature, attaining a maximum at the

optimum temperature, which lies between 33° and 36° C.; the rate then undergoes a slow depression. At higher temperatures, such as 50° C., the plant becomes limp and disorganised.

The plant has been accustomed to the normal temperature, 30° C., of its environment. We next attempted to study the effect of lower temperatures than this normal.

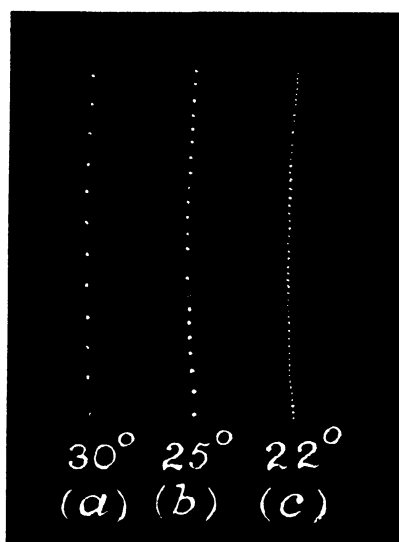


FIG. 18. Effect of temperatures below normal on growth of the leaf of *Vallisneria spiralis*.

- (a) Record of normal rate.
- (b) Record of effect of temperature at 25° C.
- (c) Record of effect of temperature at 22° C.

Experiment 3. *Effect of lower temperatures than the normal.*—After taking the record of normal growth at 30° C., cold water from the reservoir was allowed to flow into the outer vessel and thus lower the temperature of the inner chamber to 25° C., 22° C., and 18° C. successively. The records (fig. 18) show that at 30° C. it is 11, at 25° C. it is 22, and at 22° C. it is 57. In this way it was found that the rates of growth underwent continuous depression until there

was an arrest at 18° C. ; when the temperature was raised again to 20° C. there was a revival of growth. The minimum temperature for arrest of growth may therefore be taken as the mean of 20° C. and 18° C., that is 19° C.

The summary of results of variation induced in the rates of growth of the water plant *Vallisneria* between the temperatures 19° C. and 45° C. is given in the following table.

TABLE I.—RATES OF GROWTH AT DIFFERENT TEMPERATURES OF LEAF OF WATER PLANT (*VALLISNERIA*).

Temperature	Absolute rate of growth in μ per second					
19° C.	Arrest
22° C.	0.02
25° C.	0.05
30° C.	0.10
33° C.	0.12
36° C.	0.12
40° C.	0.10
45° C.	0.092

It is thus found that there is a critical minimum temperature of about 19° C., below which the growth of the water plant is arrested. The rate of growth at first increases slowly and then faster with the rise of temperature, attaining a maximum rate between 33° C. and 36° C. Above this maximum temperature the rate diminishes slowly up to 45° C. At 50° C. and above the plant becomes disintegrated.

The curve (fig. 19) gives a graphic representation of the variation in the rate of growth with changes of temperature.

SUMMARY

Investigations on the effect of temperature in inducing variation in the rate of growth of a water plant were carried out with the leaf of *Vallisneria spiralis*.

Special methods were devised for rapid and accurate determination of variation in the rate of growth under changes of temperature above and below the normal.

Automatic records for this purpose were obtained with the *High Magnification Crescograph*.

It has thus been found that under the environmental conditions to which the plant has been accustomed the rate of growth below the normal undergoes a continuous diminu-

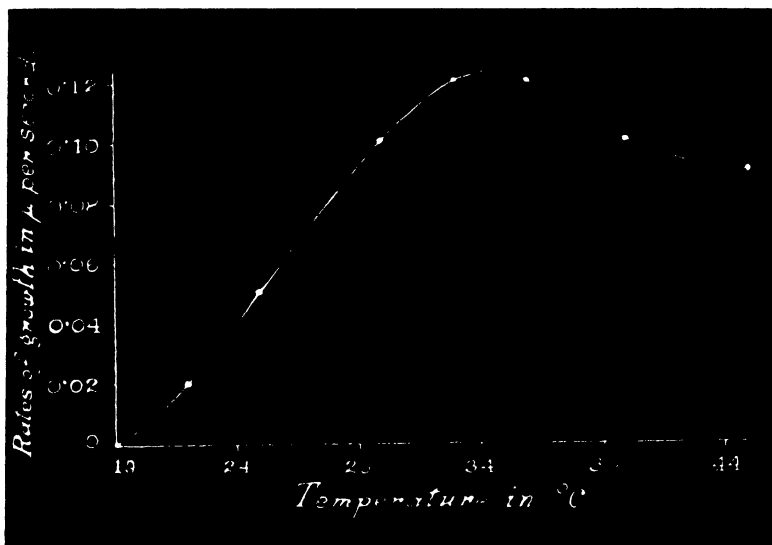


FIG. 19 Graphic representation of the relation between the rates of growth and temperature

tion, till at the critical minimum temperature of about 19° C. there is an arrest of growth.

Under rise of temperature above the normal 30° C. there is an enhancement in the rate of growth, the optimum rate being attained between 33° C. and 36° C. Above this optimum there is a slow diminution in the rate of growth. At 50° C. the plant becomes quite limp and the tissue becomes disintegrated.

IV.—INVESTIGATION ON THE OXYGEN CONSUMPTION OF THE SEED OF *CAJANUS*

BY

A. GUHA THAKURTA, C.S.AG.C., AND B. K. DUTT, B.Sc.

IN our previous paper on the 'After-ripening' of the seed of *Cajanus*, published in the *Bose Inst. Trans.*, vol. x, it was found that the seed in the resting stage is not capable of absorbing water and that the moisture percentage in the pre-resting seed has an important effect on the time of germination. The present work was undertaken partly with a view to investigating the behaviour of the resting seed as regards the absorption of atmospheric oxygen and also with the object of obtaining data which would establish the nature of the relationship between the oxygen consumption and the percentage of moisture in the seed, both in the pre- and post-resting stages.

In order to carry out a complete investigation of the oxygen consumption of the seed of *Cajanus*, numerous experiments were conducted with seeds at all the different stages: (1) Pre-resting, (2) Resting, and (3) Post-resting. Later on, a few experiments were also conducted with post-resting seeds soaked in water to investigate the relationship of oxygen consumption with the water content of the seed in that condition.

METHOD EMPLOYED

In the present investigation the oxygen consumption was in most cases recorded by the Automatic Respirograph, the

detailed description of which has been published in our previous communication.¹ When the process of oxygen consumption was extremely slow, necessitating a long time to determine the rate, this was measured by the manometric method. In this method the seeds were enclosed in a sealed chamber containing soda-lime and fitted with a mercury manometer for observing the change of pressure due to the consumption of oxygen. The advantage of soda-lime over sodium hydroxide is that it absorbs the moisture along with CO_2 inside the chamber, thereby preventing the growth of fungi during long-continued observation. The manometric change for the first 24 hours was not taken into account to obviate the error due to the absorption of water vapour and of CO_2 originally contained in the chamber. In measuring the consumption of oxygen the mercury levels of the manometer were, therefore, taken into account after the expiration of this period. The reduction of pressure which was due to the consumption of oxygen by the respiring material was noted in the manometer. For the determination of the volume of oxygen consumed a graduated burette filled with mercury was connected with the respiration chamber and mercury was dropped out slowly till there was a reduction of pressure inside the chamber which corresponded to that previously noted in the manometer. The volume of mercury dropped out of the burette thus represented the volume of oxygen consumed during the period of observation. The rate of oxygen consumption is expressed in terms of c. mm. per minute per gram of dry and fresh weights of the respiring material.

In conducting experiments with pre-resting seeds they were first divided into groups, with special care that all the members in a group were similar in maturity. Similarity amongst the individual members of each group was judged from external appearance such as size, shape and colour.

¹ A. Guha Thakurta and B. K. Dutt, 'An Automatic Respirograph,' *Trans. Bose Research Inst.*, vol. ix, pp. 77-88 (1933-34).

In obtaining resting seeds, semi-dried pods were collected. The seeds after separation from the pods were divided into groups according to their external appearance. These seeds had not yet attained the resting stage; they were brought to that condition by drying in the sun. The different groups thus brought to the resting stage were separately stored in bottles. Though the seeds in all of these groups had attained the resting stage the water content of the groups was different. Therefore, in conducting experiments in the resting and post-resting conditions, each of these groups was separately dealt with.

The moisture percentage and the rate of oxygen consumption of each individual group were determined from the same seeds, which were dehydrated after the rate of their oxygen consumption had been recorded. In seeds of very low moisture content, however, the moisture percentage and oxygen consumption of each group were determined from different quantities of the same group.

In determining the rate of oxygen consumption the seeds were not supplied with any water inside the respiratory chamber. Thus the seeds in the chamber had no chance of absorbing water and respiration depended absolutely on their internal moisture condition.

In determining the rate of respiration of post-resting soaked seeds, their surface was dried before they were introduced into the respiratory chamber.

Under the manometric method, when soda-lime was used in the respiratory chamber, the seeds were weighed twice, both before the introduction into the chamber and also after they were taken out, as some of the moisture content of the seed was found to have been lost during the period of prolonged observation. The rate of oxygen consumption was calculated in relation to the mean value of the two weights.

TABLE I.—RATE OF OXYGEN CONSUMPTION OF THE PRE-RESTING SEEDS OF *CAJANUS* HAVING DIFFERENT MOISTURE PERCENTAGES.

Moisture content of the seed	Rate of oxygen consumption per gram of the fresh weight per minute	Rate of oxygen consumption per gram of the dry weight per minute
per cent.	c.mm.	c.mm.
80.5	89.0	456.7
79.4	85.01	418.7
79.105	83.1	398.3
78.0	78.12	358.2
66.1	15.2	44.8
65.5	14.01	40.6
64.9	13.0	37.0
58.8	10.1	24.5
52.422	7.01	14.6
48.9	5.02	9.68
42.032	4.12	7.107
37.906	3.05	4.91
25.3	1.7	2.27
20.12	0.51	0.63
16.03	0.15	0.179
14.015	0.03	0.034
12.01	0.00	0.0053

OXYGEN CONSUMPTION AND THE MOISTURE CONTENT OF THE PRE-RESTING SEED

We have previously shown¹ that the resting stage is attained in the seed of *Cajanus* when its moisture percentage falls down to the neighbourhood of 10. In studying the oxygen consumption in the pre-resting condition, therefore, seeds of varying moisture content above this were separately experimented upon. The results of the experiments are summarised in Table I. The rate of oxygen consumption has been calculated in relation to both the fresh and the dry weights.

It will be evident from the data contained in Table I that the oxygen consumption of the pre-resting seed of *Cajanus* undergoes a continuous decrease as the moisture percentage falls down gradually from 80 to 12. The rate of oxygen consumption is very high at about 80 per cent. of moisture content. There is a rapid decline of the consumption of oxygen from 80.5 to 66.1 per cent. of moisture content. Afterwards the rate of fall is progressively lowered to 12.01 per cent. of moisture content. The relation between the oxygen consumption and the moisture content of the pre-resting seed is graphically represented in fig. 20.

THE OXYGEN CONSUMPTION OF THE SEED IN THE RESTING STAGE

The cause of dormancy in certain seeds has been found to be associated with the restricted absorption of oxygen by the embryo. In investigating the cause of dormancy of the seed of *Ambrosia trifida*, Davis² concluded that the embryo of *Trifida* seed is enclosed by a nucellar membrane, the low

¹ B. K. Dutt and A. Guha Thakurta, 'Investigation on the After-ripening of the Seed (*Cajanus*),' *Trans. Bose Research Inst.*, vol. x, pp. 73-91 (1935-36).

² W. E. Davis, 'After-ripening and the Development of Secondary Dormancy in the Embryo of *Ambrosia trifida*,' *Amer. Jour. of Bot.*, vol. 17, pp. 58-78 (1930).

permeability of which restricts gaseous exchange; hence, according to him, the dormancy of the seed is associated

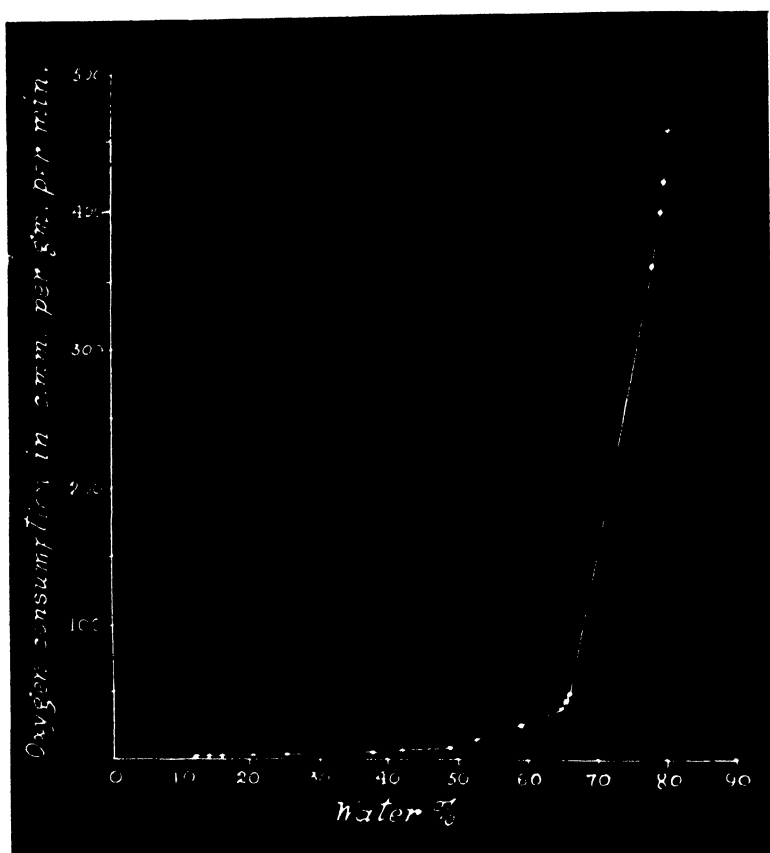


FIG. 20. Curve showing the relationship between the oxygen consumption and moisture content of the pre-resting seed of *Cajanus*.

with the restricted respiration. From the works of Crocker ¹ and Shull ² it would appear that the delayed germination of

¹ Wm. Crocker, 'Role of the Seed Coats in delayed Germination,' *Bot. Gaz.*, vol. 42, pp. 265-91 (1906).

² C. A. Shull, 'The Oxygen Minimum and Germination of Xanthium Seeds,' *Bot. Gaz.*, vol. 52, pp. 453-77 (1911).

the upper seeds of *Xanthium* is due to certain seed-coat and embryonic characteristics associated with the supply and need of oxygen. The low permeability of the seed-coat of the upper seed to oxygen and the high oxygen demand of the embryo of the same seed for germination act, according to them, in conjunction in the delayed germination of these seeds.

Seeds of *Trifida* and *Xanthium*, though incapable of germination due to the restricted absorption of oxygen, can, however, absorb water and swell in the dormant stage. But the testa of the resting seed of *Cajanus*, as we have shown previously,¹ is absolutely impermeable to water. The object of the present investigation was, therefore, to determine whether the impermeability of the testa of the resting seed of *Cajanus* is limited to the power of absorption of water alone or if it is also due to the inability of absorbing oxygen as well.

It has been previously stated that in obtaining seeds in the resting condition they were collected just before the resting stage was attained ; they were then dried for attaining the resting stage. Thus the exact time when the resting stage was initiated was known. Since the seeds do not ripen all at the same time, several lots of resting seeds were collected at different times of the season.

After finding that the resting stage had been attained, the seeds were enclosed in the respiration chamber with the manometric device. The seeds under this condition were found not to exhibit any sign of absorbing oxygen for a considerable length of time. In most of the experiments the observation was limited for about a month ; in none of them was the resting seed found to absorb oxygen. Some of the experiments were continued for a longer time, till the period of rest was over. In those experiments the internal pressure of the respiration chamber was found to decrease at a very

¹ B. K. Dutt and A. Guha Thakurta, 'Investigation on the After-ripening of the Seed (*Cajanus*),' *Trans. Bose Research Inst.*, vol. x, pp. 73-91 (1935-36).

slow rate after a time which corresponded approximately with the period of rest of the seed. The results of two typical experiments in which the observation was continued even after the termination of the approximate resting period are given in Table II.

TABLE II.—THE OXYGEN CONSUMPTION OF THE SEED OF *CAJANUS* JUST AFTER THE RESTING STAGE WAS TERMINATED.

Exp.	Moisture percentage of the seed	Approximate period of rest within which no manometric fall could be detected	Rate of oxygen consumption just after the resting stage was terminated with relation to fresh weight per gram per minute	Rate of oxygen consumption just after the resting stage was terminated with relation to dry weight per gram per minute
	per cent.	days	c.mm.	c.mm.
	8.72	57	0.00051	0.00056
	7.02	90	0.00044	0.00048

The average oxygen consumption was deduced from the total consumption of 12 days in Experiment 1 and 10 days in Experiment 2, after the first manometric fall was detected.

THE OXYGEN CONSUMPTION IN THE POST-RESTING CONDITION

It was noticed that the seeds which in the resting stage cannot absorb any moisture, having been steeped in water for a long time, became partially hygroscopic in the post-resting condition. The bottles in which the seeds were stored were not airtight and the post-resting seeds under this condition were found to increase gradually in their water percentage. The rate of oxygen consumption, which was so feeble at the termination of the resting stage, was also

found to increase with the increasing water content of the post-resting seeds. This will be clear from the accounts of two observations given in Table III.

TABLE III.—INCREASE OF OXYGEN CONSUMPTION OF THE STORED SEED OF *CAJANUS* DUE TO ABSORPTION OF ATMOSPHERIC MOISTURE AFTER THE CESSATION OF RESTING PERIOD.

Group No.	Moisture content of the post-resting seed	Rate of oxygen consumption per gram of the fresh weight per minute	Rate of oxygen consumption per gram of the dry weight per minute
	8.72 %	0.00051	0.00056
	11.01 %	0.0013	0.0015
	7.02 %	0.00044	0.00048
	9.5 %	0.001	0.0011

In Group 1 the moisture percentage increased from 8.72 to 11.01 in about 54 days, between April 18 and June 12. In Group 2 the moisture percentage increased from 7.02 to 9.5 in about 43 days, between June 8 and July 21. Group 2 is therefore seen to have absorbed more moisture in a comparatively shorter period. This might be due to the higher moisture content of the atmosphere during that period.

THE OXYGEN CONSUMPTION OF THE POST-RESTING SEED SOAKED IN WATER

After finding that the oxygen consumption of the post-resting seed increases with the increase of moisture content of the seed absorbed from the atmosphere, we next tried to

investigate the modification in the rate of oxygen consumption of the post-resting seed having different moisture content due to direct absorption of water. The investigation was undertaken in two different ways: (1) by soaking different quantities of seeds for different periods and thereby obtaining partially soaked seeds of varying moisture content; (2) by soaking a certain quantity of seeds to the point of germination and then gradually bringing them down to lower moisture content by dehydration. The rates of oxygen consumption at different moisture conditions of the seed were determined side by side with the determination of moisture content.

Method 1.—For obtaining partially soaked seeds of varying moisture content from very low to higher values, separate lots from the same group were allowed to absorb water for different periods, from 1 to 8 hours. After the seeds were taken out of the water they were surface-dried by carefully rolling them on dry filter-paper and afterwards keeping them under an electric fan for a certain length of time. After this mode of partial soaking, the swelling of the seed is not uniform, but occurs at different places, causing irregular protuberances on the surface. But when such a seed is surface-dried and allowed to remain in that condition for several hours, the absorbed moisture is uniformly distributed all over the seed, causing regular expansion. The seeds were then weighed in this condition and put into the respiration chamber. The original moisture percentage and the dry matter content of the seed being known, the actual moisture content at the soaked state was easily deduced from the weight of the soaked seed. The rate of oxygen consumption at such moisture condition of the seed was expressed on the basis of dry and soaked weights of the seed. From the variation in the time of soaking in water it was thus possible to obtain seeds of different moisture content and study the oxygen consumption under those conditions. The results of a few experiments are given in Table IV.

TABLE IV.—OXYGEN CONSUMPTION OF THE POST-RESTING SEEDS OF *CAJANUS* PARTIALLY SOAKED IN WATER. (THE ORIGINAL MOISTURE CONTENT OF THE SEED WAS 9.5 %.)

Number of hours soaked	Moisture percentage of the surface-dried seeds	Rate of oxygen consumption per gram of the soaked weight per minute	Rate of oxygen consumption per gram of the dry weight per minute
1	per cent. 12.1	c.mm. 0.0035	c.mm. 0.0043
2	16.32	0.0078	0.012
3	19.7	0.016	0.021
4	24.58	0.0402	0.051
5	28.33	0.081	0.105
6	34.52	0.22	0.308
7	38.93	0.607	0.854
8	44.52	1.39	2.52

Method 2.—In soaking the seeds to the point of germination they were soaked in water for about 17 hours. The seeds in this condition became fully swollen, some of them even burst. The seeds under this condition were taken out of the water and surface-dried by the process already described. After taking the weight of the surface-dried seeds to find out the moisture content, the rate of their oxygen consumption

was determined. After that the seeds were kept exposed in the sun for some time for partial dehydration, cooled down under an electric fan and then the moisture content and the rate of oxygen consumption were determined under that condition. In this way the same seeds were gradually brought down to lower moisture content and rates of oxygen consumption were determined under those conditions. The results of two series of experiments are given separately in Tables V and VI.

TABLE V.—THE RATE OF OXYGEN CONSUMPTION AT DIFFERENT MOISTURE CONTENTS OF THE POST-RESTING SEED OF *CAJANUS* WHEN GRADUALLY DRIED FROM FULLY SOAKED CONDITION. (THE MOISTURE CONTENT OF THE DRY SEED BEFORE SOAKING WAS 11.34 %)

Water content of the soaked seed	Rate of oxygen consumption per gram of the fresh weight per minute	Rate of oxygen consumption per gram of the dry weight per minute
per cent.	c.mm.	c.mm.
55.0	4.2	9.2
47.8	3.8	7.3
31.2	0.46	0.67
20.69	0.092	0.12
7.9	0.011	0.012

It will be evident from the comparison of data contained in Table IV with those of Tables V or VI that the fully soaked seeds when brought down to lower moisture condition consume more oxygen than the partially soaked seeds of the

TABLE VI.—THE RATE OF OXYGEN CONSUMPTION AT DIFFERENT MOISTURE CONTENTS OF THE POST-RESTING SEED OF *CAJANUS* WHEN GRADUALLY DRIED FROM FULLY SOAKED CONDITION. (THE MOISTURE CONTENT OF THE DRY SEED BEFORE SOAKING WAS 9.5 %.)

Water content of the soaked seed	Rate of oxygen consumption per gram of the fresh weight per minute	Rate of oxygen consumption per gram of the dry weight per minute
per cent. 54.8	c.mm. 4.0	c.mm. 8.84
47.0	3.22	6.07
44.7	2.75	4.97
41.3	2.26	3.85
38.6	1.69	2.75
35.98	1.06	1.65
30.1	0.38	0.543
16.9	0.042	0.0504
6.1	0.0076	0.008

corresponding moisture content. The cause of this may be attributed to the growth of certain enzymes in the fully soaked germinating seeds, the activity of which is partially retained even when the seeds are brought down to lower moisture content.

The data of Tables IV and VI are given in curves *a* and *b*, in fig. 21, to show more clearly the comparative difference

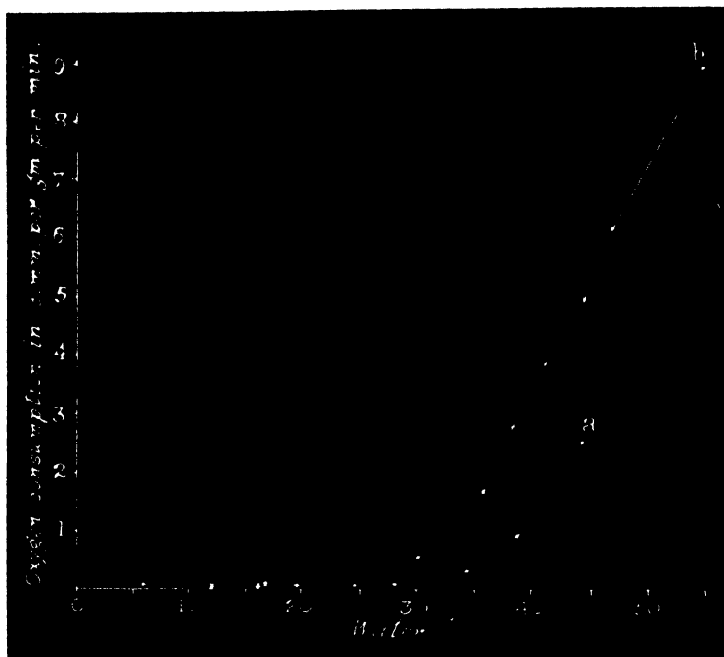


FIG. 21. Curves showing the relationship between the oxygen consumption and moisture content of the post-resting soaked seed of *Cajanus*.

- a. Represents the oxygen consumption of the seed at partially soaked condition.
- b. Represents the oxygen consumption of the seed when brought down gradually to lower moisture contents from fully soaked condition.

of oxygen consumption at similar moisture content, attained under different treatments as stated above.

SUMMARY

Investigations on the oxygen consumption of the seeds of *Cajanus* were undertaken to find out whether in the resting state they were capable of absorbing oxygen.

Attempt was also made to find out the relation between the oxygen consumption and the moisture percentage of the seed both in pre- and post-resting stages.

It has been found that the oxygen consumption of the pre-resting seed is continuously decreased along with the decrease of moisture percentage of the seed. The rate of oxygen consumption at about 80 per cent. of water content is very high. From about 80 per cent. to 66 per cent. the fall of the rate of oxygen consumption is very rapid, and after that the rate of fall is progressively lowered to about 12 per cent. of moisture content.

It has also been found that in the resting stage the oxygen consumption is absolutely nil. When the resting stage is passed the post-resting seed begins to absorb oxygen very slowly; the absorption of oxygen, however, increases when the moisture content of the post-resting seed is increased by absorption of water from the atmosphere.

The oxygen consumption of partially soaked post-resting seed is progressively increased along with the increase of water content of the seed. When the fully soaked post-resting seed is gradually brought down to lower moisture content by dehydration, the oxygen consumption is gradually decreased. The consumption of oxygen under this condition is, however, comparatively greater than that of the partially soaked seed of corresponding moisture content.

ADDITIONAL BIBLIOGRAPHY

- (1) W. McK. Atwood: 'A Physiological Study of the Germination of *Avena fatua*,' *Bot. Gaz.*, vol. 57, pp. 386-414 (1914).
- (2) Wm. Crocker: 'Mechanics of Dormancy in Seeds,' *Amer. Jour. Bot.*, vol. 3, pp. 99-120 (1916).
- (3) D. A. Coleman, B. E. Rothgeb and C. H. Fellows: 'Respiration of *Sorghum* Grains,' *U.S. Dept. Agr. Tech. Bull.* 100 (1928).
- (4) C. H. Bailey and A. M. Gurjar: 'Respiration of Stored Wheat,' *Jour. Agr. Res.*, vol. 12, pp. 685-713 (1918).
- (5) Jean White: 'The Ferments and Latent Life of Resting Seeds,' *Proc. Roy. Soc., B*, vol. 81, pp. 417-442 (1909).
- (6) R. Jacquot and A. Mayer: 'Hydratation et respiration des graines,' *Ann. Physiol. Physiochem. Biol.*, vol. 2, pp. 408-425 (1926).

V.—INVESTIGATION ON PHYSIOLOGICAL REACTIONS OF PLANTS UNDER LOW ATMOSPHERIC PRESSURE

BY

B. K. DUTT, B.Sc., AND A. GUHA THAKURTA, C.S.AG.C.

THE study of the effects of low atmospheric pressure on man and animal was first undertaken by Paul Bert ¹ as early as 1878, and his experiments conclusively showed that the physiological effects produced by low atmospheric pressure are not simply due to the diminished mechanical pressure but are the results of diminished partial pressure of oxygen. The idea that oxygen could be used to avert the very dangerous effects of rarefied air in balloon ascents was also discovered by Paul Bert.

Extensive investigation on this line was also carried out by Haldane,² who fully supported the reasonings of Paul Bert. He found that the reduction of external pressure profoundly affected the pulsations of the heart of man. The question then arose whether this was due to the effect of diminution of pressure as such or due to the diminution of partial pressure of oxygen in the atmosphere.

From the results of his experiments Haldane came to the conclusion that the change in the heart-beat of man under diminished pressure is due to the reduction of partial pressure of oxygen; for when this partial pressure of oxygen was kept the same, a diminution of atmospheric pressure did not produce any change in the normal rate of heart-beat.

¹ Paul Bert, *La Pression Barométrique* (1878).

² J. S. Haldane and J. G. Priestley, *Respiration* (1935).

EFFECT OF VARIATION OF EXTERNAL PRESSURE ON THE PULSATORY ACTIVITY OF PLANTS

The researches that have been carried out in the Bose Research Institute¹ have shown that the pulsatory mechanism in the plant is essentially similar to that in the animal, both being similarly affected by external agencies. We thought it would be of very great interest to find whether variation of external pressure affected the pulsatory activity of the plant. If so, whether the effect was in any way due to the reduction of partial pressure of oxygen present in the atmosphere.

METHOD OF EXPERIMENTAL INVESTIGATION

For experimentally producing low atmospheric pressure, a vacuum desiccator and an electrically-driven rotary suction pump were employed. A graduated mercury manometer was attached to the desiccator to indicate the pressure. The desiccator was connected with the pump through a glass stop-cock. After a definite reduction of pressure was produced in the desiccator, the stop-cock was closed and the same pressure was maintained for a definite period necessary for the experiment. Another glass tube for introducing oxygen reached to the bottom of the chamber and was connected to the oxygen reservoir through another stop-cock.

The observation was made by directly recording the movements of the pulsating plant organ. The apparatus employed for recording was a *Single Lever Oscillating Recorder*, specially constructed in the Institute for the present purpose. The lateral movement of the recording plate lasted for 75 minutes, which was the maximum time-limit of each experiment. The dots were at intervals of 10 seconds. The plant was attached to the lever of the recorder by a fine silk thread, after which the recorder with

¹ Sir J. C. Bose, *The Motor Mechanism of Plants*, pp. 240-73 (1928).

the mounted plant was introduced into the desiccator. The experimental arrangement is diagrammatically represented in fig. 22.

For the pulsating organ in the plant a leaflet of *Desmodium gyrans* was employed, a cut-specimen being utilised for the

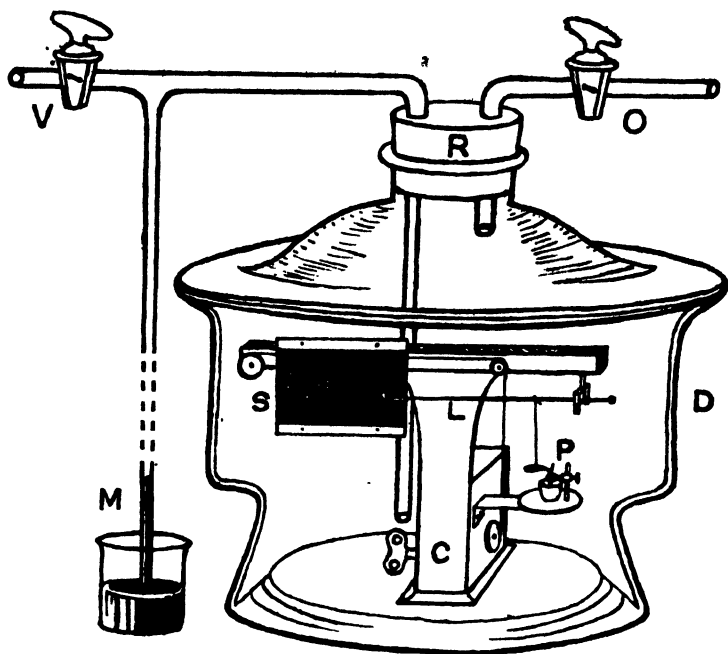


FIG. 22. Experimental arrangement for observing the effect of reduced atmospheric pressure on plant.

- D, desiccator used as plant chamber. R, rubber stopper. Air inside the chamber is pumped out through the glass stop-cock, V. M, mercury manometer for observing the pressure inside the chamber. O, glass stop-cock for introducing oxygen. P, pulsating leaflet of *Desmodium* attached to the recording lever, L. C, clockwork arrangement for to-and-fro oscillation and lateral movement of the smoked plate, S.

purpose. Care was, however, taken to allow sufficient time for the disappearance of the irritation caused by the wound, the normal rate of pulsation being thus restored. The recorder with the attached leaflet was then enclosed in the chamber. Three or four pulses were recorded under normal

pressure, after which the pressure was diminished to a certain value ; this was maintained constant and its effect on pulsation was recorded. In this way the effects of diminished external pressure ranging from 200 mm. to 600 mm. below the normal were separately recorded on different specimens. Each experiment was repeated several times on a number of specimens for verification of the effect of a given pressure. It should be mentioned here that the

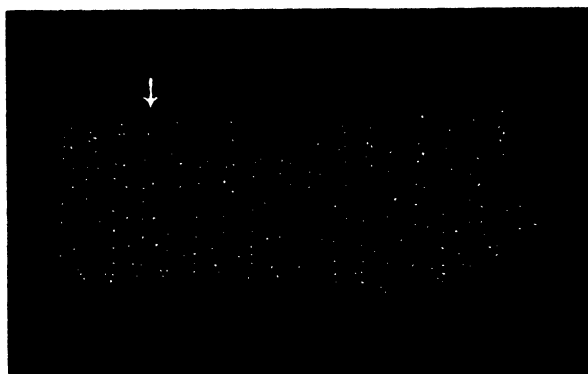


FIG. 23. Effect of reduced pressure of 560 mm. (200 mm. below normal).

The last seven pulses are seen to be irregular and increased in frequency.

results of the different experiments at each particular pressure were quite in agreement with one another. Typical records of effects of variation of pressure on autonomous pulsation of *Desmodium* are given in figs. 23, 24, 25 and 26. Reduction of pressure is marked with an arrow ; when the pressure is restored to normal it is indicated by W and arrow. Below is given the detailed description of a number of typical experiments.

Experiment 1.—At a pressure of 560 mm., or 200 mm. below normal (fig. 23), pulsations after diminution of pressure at arrow were at first uniform and similar to those at normal ; but the last seven pulses became irregular and

slightly increased in frequency as calculated from the number of dots in each pulse. The average number of dots in the last seven pulses is only 20 as against the normal average of 23.

Experiment 2.—At 460 mm., or 300 mm. below normal, the pulses became irregular, being definitely increased in frequency immediately after the reduction of pressure (fig. 24). The average number of dots under reduced pressure is 23 against the normal average of 27, showing that there is an increase of frequency of about 14 per cent.

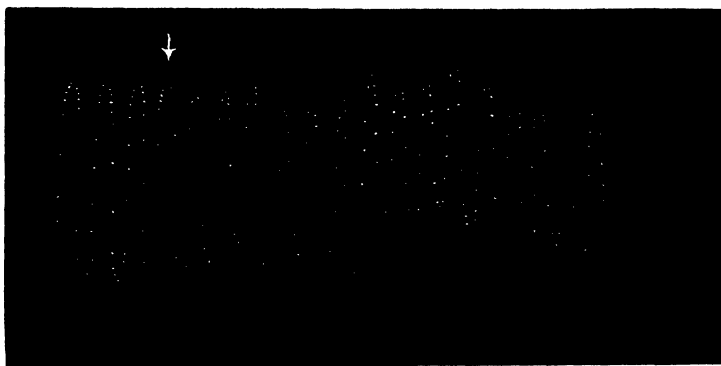


FIG. 24. Effect of reduced pressure of 460 mm. (300 mm. below normal).

The record shows the increase of frequency of pulsation immediately after the reduction of pressure.

Experiment 3.—At 360 mm., or 400 mm. below normal, the amplitude of the pulses became immediately reduced, culminating practically in an arrest (fig. 25). After restoration of the normal pressure at W the pulsations underwent a quick recovery. The last three pulses after the restoration of normal pressure are seen to have regained almost their normal amplitude and frequency.

Experiment 4.—The reduced pressure of 260 mm., or 500 mm. below normal, brought about an immediate arrest of the pulsatory activity (fig. 26). The specimen was subjected to the reduced pressure for about 20 minutes, after

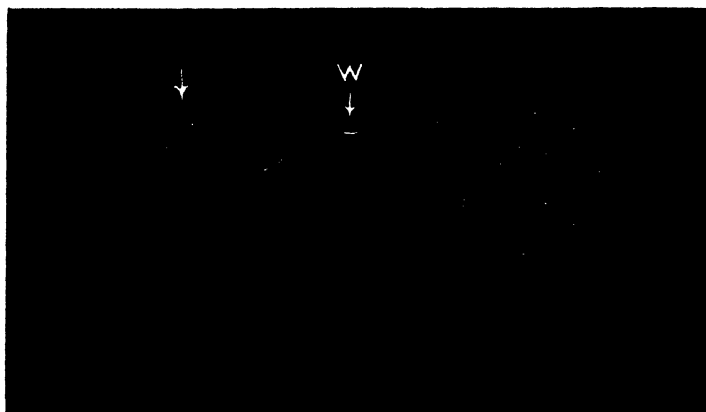


FIG. 25. Effect of reduced pressure of 360 mm. (400 mm. below normal) and of subsequent restoration of normal pressure.

The record shows arrest of pulsation and gradual recovery on return to normal pressure.

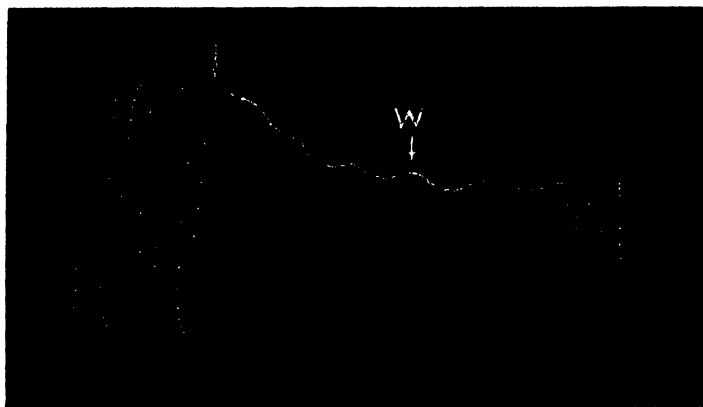


FIG. 26. Effect of reduced pressure of 260 mm. (500 mm. below normal) and of subsequent restoration of normal pressure.

The record shows complete abolition of pulsation and partial recovery on return to normal pressure.

which the normal pressure was restored. Partial recovery was initiated after 15 minutes of restoration of normal pressure, complete recovery occurring after 90 minutes.

In some experiments the pressure was lowered to 160 mm., or 600 mm. below normal; the pulsation stopped immediately after the reduction of pressure. The pulsatory activity did not, however, return till several hours after the restoration of normal pressure.

In the case of the animal heart it has been stated that the irregularity in its pulsations brought about by diminution of external pressure is really due to the diminished partial pressure of oxygen. For even under diminished external pressure, if the partial pressure of oxygen is maintained equal to that under normal pressure, the pulsatory activity of the heart undergoes no variation. .

In regard to the parallel phenomenon of pulsatory activity in the plant, is the variation induced under diminished pressure due to the change of pressure as such, or is it brought about by diminution of partial pressure of oxygen? The only way of obtaining a satisfactory answer to this question is by observing the effect of diminished external pressure, utilising special means of maintaining the partial pressure of oxygen constant.

Many experiments were carried out along this line. The leaflet of *Desmodium* was at first subjected to diminished external pressure, say of 260 mm., or 500 mm. below normal. In regard to the effect of purely diminished external pressure, reference to fig. 26 will show that it causes an immediate arrest of pulsatory activity.

In the present series of experiments we tried to find out whether maintenance of partial pressure of oxygen as under the normal pressure of 760 mm. rendered the pulsatory activity of the plant immune to the effect of diminution of external pressure.

A detailed account of a typical experiment will make clear the procedure adopted.

Experiment 5.—The pressure in the chamber was, after a series of operations, reduced to 260 mm., or 500 mm. below normal. As previously stated, reference to fig. 26 shows that this reduction of pressure induces an arrest of pulsation. But in the present case care was taken that the partial pressure of oxygen was maintained equal to that under normal pressure. At the above diminished pressure the partial pressure of oxygen is 52 mm. But in order to maintain the partial pressure of oxygen equal to that at the normal external pressure of 760 mm., namely of 152 mm.,

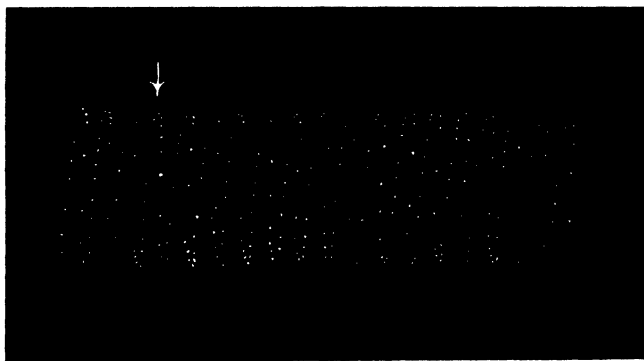


FIG. 27. Reduced pressure of 260 mm. (500 mm. below normal) has no effect when the partial pressure of oxygen is maintained equal to that of the normal atmosphere.

the partial pressure was carefully raised to this value by introducing requisite quantities of oxygen into the plant-chamber. The result was that the pulsatory activity remained unaffected, as shown in fig. 27.

From the results of the experiments it can be concluded that the effect of reduced pressure on the pulsatory activity of *Desmodium* is only due to the fall of the partial pressure of oxygen. When the partial pressure of oxygen is maintained equal to that of the normal atmosphere, the pulsatory activity remains unaltered even at a very low pressure which would otherwise have arrested it.

EFFECT OF VARIATION OF EXTERNAL PRESSURE ON MOTO-EXCITABILITY OF PLANT

Haldane and others have found that the muscular co-ordination is affected at a low pressure. The present investigation was undertaken to determine the effect of low pressure on the contractile tissue of the plant which has a similar function to that of the muscular tissue of an animal.

For researches of moto-excitability of plants the pulvinar tissue of *Mimosa pudica* has been found to be most suitable. The work carried out in the Institute has shown ¹ that the excitability of the plant tissue under minimally effective electric stimuli, as indicated by the contractile response of the pulvinus, remains fairly constant for a long period, the external condition being constant. Investigation was therefore undertaken to find out how the moto-excitability of the pulvinus of *Mimosa* is affected under reduced external pressure.

METHOD OF EXPERIMENT

The experiments were performed with intact potted plants. The second or third leaf from the apex was, generally speaking, selected for the experiments. The record was taken with a *Single Lever Oscillating Recorder* similar to that employed for recording the pulsation of *Desmodium*. The petiole of the leaf was attached by a fine silk thread to the lever, so that the fall of the leaf caused by the contraction of the pulvinus was recorded as a down-movement. The movement of the recording plate lasted for 75 minutes, which was the maximum time limit for each experiment. The dots in the records were at intervals of 30 seconds.

The pulvinus was directly excited by tetanising electric shock from an induction coil, the secondary terminals

¹ Sir J. C. Bose, *The Motor Mechanism of Plants*, pp. 90-105 (1928).

of which were connected with the plant inside the chamber through two holes in the rubber stopper of the desiccator. The intensity of the shock could be increased from the minimal to the maximal by moving the secondary coil nearer the primary. The duration of the shock was adjusted by interposing a metronome in the primary circuit of the

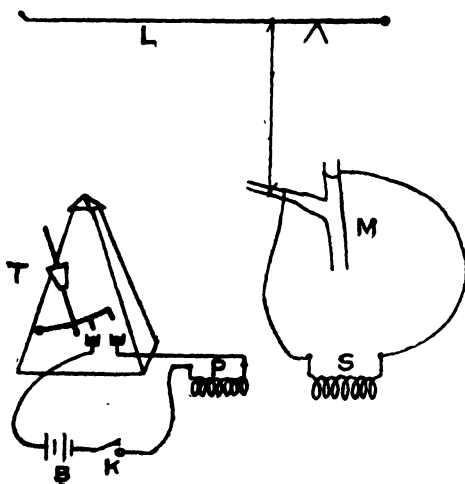


FIG. 28. Diagrammatic representation of the electric circuit for excitation of the pulvinus of *Mimosa*.

The petiole of *Mimosa* (M) is attached by a silk thread to the lever L. The pulvinus is directly stimulated through the secondary circuit S. P, primary coil connected in series with tapping key K, battery B, and metronome T.

induction coil. The electric circuit is diagrammatically represented in fig. 28.

Uniform shocks of minimally effective intensity were applied at intervals of 15 minutes. As the maximum time-limit of our experiment was 75 minutes, 5 responses were recorded on each plate. Two responses were thus recorded under normal pressure; the pressure was then lowered to a certain definite value and its effect on the responses recorded. The results obtained with different

specimens were found to be very similar. The experiments described below may be regarded as typical.

Experiment 6.—In fig. 29 the effect of reduced pressure of 460 mm. or of 300 mm. below normal has been given. After reduction of the pressure the amplitudes of the responses became slightly decreased, indicating that the excitability had undergone a slight diminution.

Experiment 7.—At 360 mm., or 400 mm. below normal, rapid diminution occurred in the amplitude of the response,

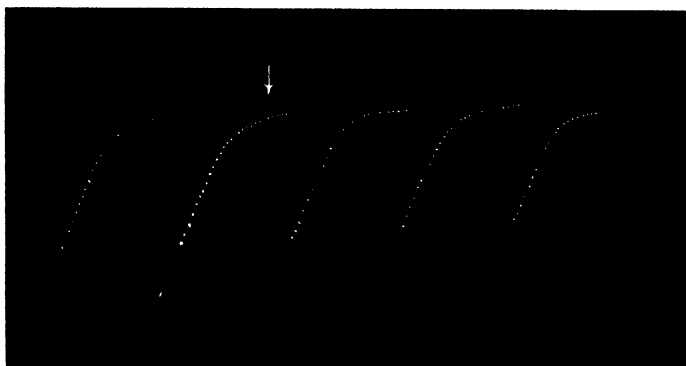


FIG. 29. The reduced pressure of 460 mm. (300 mm. below normal) induces a slight diminution of moto-excitability.

which underwent continuous diminution. After the restoration of the normal pressure at W there was, however, a complete recovery, as shown in fig. 30.

Experiment 8.—At 260 mm., or 500 mm. below normal, repeated shocks failed to evoke any response of the plant (fig. 31). The plant, however, was found to recover when the pressure was restored to normal at W.

It will be evident from the above results that the moto-excitability of the pulvinus of *Mimosa* diminishes when the atmospheric pressure is reduced to 460 mm., or 300 mm. below normal; further reduction of the excitability occurs at 360 mm., or 400 mm. below normal; the excitability is

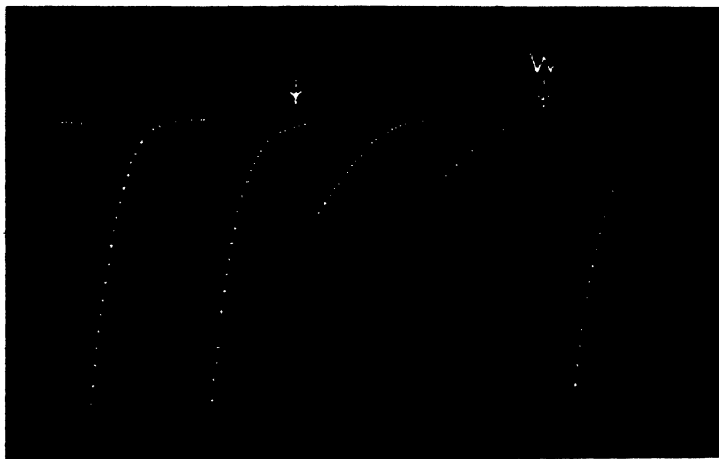


FIG. 30. Effect of reduced pressure of 360 mm. (400 mm. below normal).

The record shows diminution of excitability and full recovery on return to normal pressure.

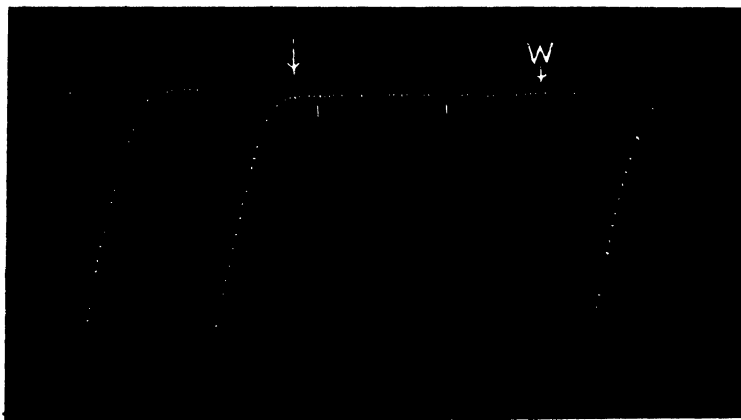


FIG. 31. Effect of reduced pressure of 260 mm. (500 mm. below normal).

Note total abolition of excitability after reduction of pressure and recovery on return to normal pressure.

completely abolished at 260 mm., or 500 mm. below normal.

Investigation was next undertaken to determine how the moto-excitability of *Mimosa* is affected by the reduction of external pressure when the partial pressure of oxygen is maintained equal to that under normal atmosphere. In doing so a few experiments were performed at a low pressure of 260 mm., or 500 mm. below normal, having

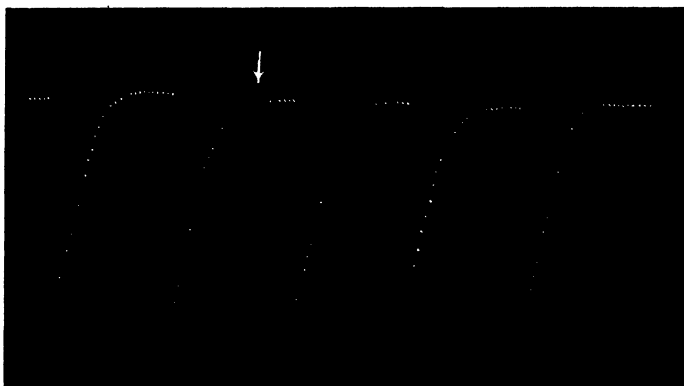


FIG. 32. Effect of reduced pressure of 260 mm. (500 mm. below normal) having the partial pressure of oxygen equal to that under normal atmosphere.

Note that the excitability remains unaffected.

the partial pressure of oxygen equal to that under normal atmosphere.

Experiment 9.—The moto-excitability of the pulvinus of *Mimosa* was found to remain quite constant even at a low pressure of 260 mm., or 500 mm. below normal, when the partial pressure of oxygen was maintained equal to that under normal atmosphere. A typical record representing the result is given in fig. 32. The first 2 responses are under normal pressure and the last 3 are under the reduced pressure of 260 mm., or 500 mm. below normal, the partial pressure of oxygen having been maintained equal to that under normal atmospheric pressure. The responses are seen to

be unaffected under the action of reduced pressure which would have normally caused total arrest of response (cf. fig. 3I).

SUMMARY

The effects of low atmospheric pressure have been studied in modifying certain physiological activities of plants, such as the Autonomous Pulsatory Activity and also the Moto-Excitability.

It has been found that the pulsation of a *Desmodium* leaflet is slightly increased in frequency at a pressure of 560 mm., or 200 mm. below normal. At 460 mm., or 300 mm. below normal, the increase of frequency is more marked. The pulsation is greatly diminished in amplitude and frequency at 360 mm., or 400 mm. below normal and becomes completely arrested when the pressure is reduced to 260 mm. or 500 mm. below normal. The normal pulsatory activity is, however, restored on return of the normal pressure.

Even under reduced pressure which arrests the pulsatory activity, the pulsations remain unaffected when the partial pressure of oxygen is maintained equal to that under normal atmospheric pressure.

In regard to the moto-excitability of plants, it undergoes a slight decrease at 460 mm. or 300 mm. below normal. The excitability becomes much diminished at 360 mm. or 400 mm. below normal and it undergoes an abolition at 260 mm. or 500 mm. below normal. After the restoration of the normal pressure there is, however, a complete recovery of the excitability. The excitability also remains unaffected even under a pressure of 260 mm. or 500 mm. below normal when the partial pressure of oxygen is maintained equal to that under normal atmosphere.

ADDITIONAL BIBLIOGRAPHY

- (1) J. S. HALDANE, A. M. KELLAS and E. L. KENNAWAY: 'Experiments on Acclimatisation to Reduced Atmospheric Pressure,' *Jour. of Physiol.*, vol. 53, pp. 181-206 (1919-20).

- (2) C. G. DOUGLAS, J. S. HALDANE, HENDERSON, YANDELL and E. C. SCHNEIDER: 'Pikes Peak Expedition,' *Phil. Trans.*, B. 203, p. 185 (1912).
- (3) H. VON SCHROTTER: 'Der Sauerstoff in der Prophylaxie und Therapie der Luftdruckerkrankungen' (1906).
- (4) MARTIN FLACK: 'The Physiological Effects of Flying,' *Nature* (London), vol. 121 (3060), pp. 986-88 (1928).
- (5) EDWARD J. VAN LIERE: 'The Influence of Anoxæmia on the Heart and Role of Pericardium in Cardiac Dilatation,' *Amer. Jour. Physiol.*, vol. 81 (2), p. 512 (1927).
- (6) N. B. DREYER: 'Some Effects of Anoxæmia on the Circulation,' *Canadian Med. Assoc. Jour.*, vol. 16 (1), pp. 26-27 (1926).

VI.—THE CHEMICAL CONSTITUTION OF *CLERODIN*

BY

H. N. BANERJEE, M.Sc.

IN a previous communication ¹ an account was given of the composition of a fixed oil obtained from *Clerodendron infortunatum*, locally known as *Bhant*, by extraction with light petroleum ether. As mentioned therein, a phytosterol (melting point 148° C.) and an alcohol (melting point 75° C.) were isolated from the extract with a very poor yield. The sterol has now been obtained in quantity. Its iodine value has been determined and found to be 124.5. It is lævo-rotatory with a specific rotation of $[\alpha]_D = -25.6$ in chloroform. From the high iodine value of this sterol, it appears probable that the compound contains two pairs of doubly linked carbon atoms. Further investigation on this sterol will be carried out when sufficient quantity of the material has been collected. Below in fig. 33 is reproduced a micro-photograph of the sterol crystals.

CLERODIN

In the same paper ¹ mention was made of a crystalline bitter principle, *Clerodin*, isolated for the first time, in a very pure form, by the author, from the leaves of *Bhant* (*Clerodendron infortunatum*). No chemical investigation seems to have been done as yet on the *Bhant* plant. The bitter substance in the leaves is supposed to possess definite

¹ H. N. Banerjee, *J. Ind. Chem. Soc.*, vol. xiv, pp. 51-57 (1937).

physiological action, the leaves being extensively used in Ayurvedic practice.

The amount of pure *Clerodin* so far collected has not been sufficient for a complete elucidation of the chemical constitution of the bitter principle. However, the investigations which form the subject-matter of this paper go to indicate the nature of the constitution of the substance.



FIG. 33. Crystals of Sterol from *Clerodendron infortunatum*.

Clerodin is a colourless crystalline substance, which when freshly prepared has a faint odour resembling *Bhant*. This smell disappears on standing. It crystallises in fine silky needles from petroleum ether and in large cubical crystals from alcohol. Fig. 34 is a representation of *Clerodin* crystals from a micro-photograph.

Clerodin is very sparingly soluble in boiling water, to which it imparts a bitter taste. In the usual organic solvents it is highly soluble even while cold. It is soluble in cold con-

centrated hydrochloric acid, nitric acid and sulphuric acid. The sulphuric acid solution is of a blood-red colour with a green fluorescence. It does not give any colour reaction or precipitate with ferric chloride, mercuric chloride, lead acetate, calcium chloride or silver nitrate. It is insoluble in dilute aqueous caustic soda solution (5 per cent.).

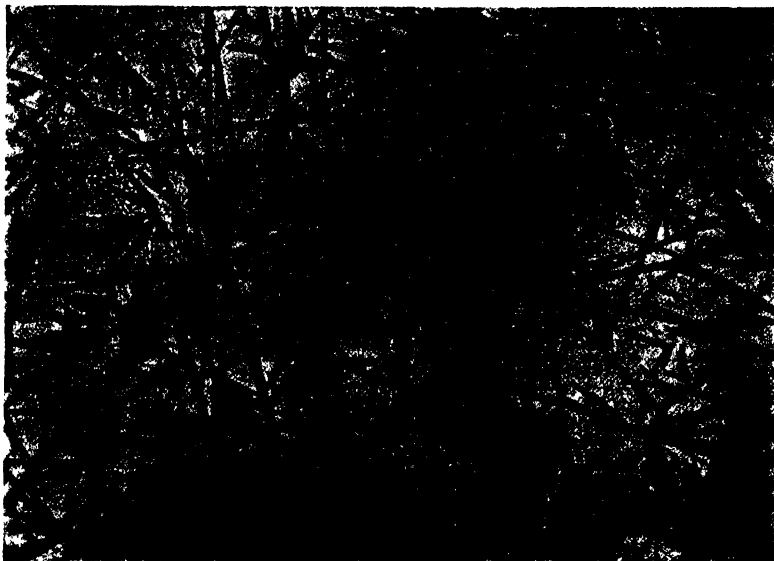


FIG. 34. Crystals of *Clerodin*.

MOLECULAR FORMULA

Results of a series of analyses and determination of molecular weight by the cryoscopic, ebullioscopic and Rast methods point to the molecular formula $C_{13}H_{18}O_3$.

One of the three oxygen atoms is in an alcoholic OH group, since *Clerodin* forms mono-acetyl, mono-benzoyl and mono-phenyl urethane derivatives.

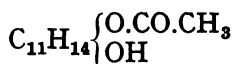
Clerodin does not react with hydroxylamine, phenyl hydrazine, or semi-carbazide, and does not reduce Fehling's solution or Tollen's reagent. It does not give any colour

reaction with alcoholic potash or with alkaline sodium nitro-prusside in pyridine solution.

Zeisel's estimation proved the absence of any methoxy group in the molecule.

Estimation by the Freudenberg¹ process revealed the presence of an acetyl-oxy group in the *Clerodin* molecule.

The three oxygen atoms of *Clerodin* are thus all accounted for, one in an alcoholic OH group, two more in an ester grouping. This relationship may be represented by the following scheme :



Clerodin is unsaturated. It decolorises bromine water slowly and bromine in chloroform quite readily. By the action of bromine in chloroform on *Clerodin* a derivative was obtained containing two bromine atoms in the molecule. Volumetric estimation of unsaturation showed the presence of one pair of double bonds in the molecule.

Clerodin is optically active, showing in chloroform solution a lævo-rotation of $[\alpha]_D = -47.6$. The optical activity and the lævo-rotation were preserved in all the derivatives of *Clerodin* prepared up to this time. The substance does not show any muta-rotation.

Heated at $105^\circ\text{--}110^\circ\text{C}$. for 4 hours, *Clerodin* did not lose any weight, nor was there any change in the crystalline shape. But when heated at $125^\circ\text{--}130^\circ\text{C}$. for 2 hours under ordinary atmospheric pressure the substance was found to decompose into a colourless vitreous mass which could not be made to crystallise.

Clerodin is a non-glucosidal bitter principle, since the substance does not yield any reducing sugar on hydrolysis. No furfural or methyl furfural is obtained from the substance on distillation with 12 per cent. hydrochloric acid. Negative Keller-Kiliani test showed the absence of any desoxy sugar in it.

¹ Freudenberg, *Annalen*, vol. 433 (1923), p. 230 ; vol. 494 (1932), p. 68.

EXPERIMENTAL

Isolation and properties.—It has already been shown that the most suitable solvent for the extraction of *Clerodin* from dry *Bhant* leaf powder is petroleum ether, which extracts the fixed and also the essential oil contained in the leaf. The proportion of *Clerodin* in the *Bhant* leaf varies within wide limits, according to locality and variety and also according to the season when the collection is made.

The purest sample of *Clerodin* prepared after several recrystallisations from alcohol (and charcoal) melts exactly at 162° C. It is a compound of C, H, and O only and does not contain N. Heated slowly on a platinum foil it first melts to a clear yellow liquid, finally charring and burning off completely with a yellow and slightly sooty flame.

Experiments performed *in vitro* with earthworms, small fishes, and worms obtained from the intestines and peritoneal cavities of fish proved that *Clerodin* is likely to act as an anthelmintic.

Combustion analysis of *Clerodin* gave the following results :

C = 70.46 per cent., 70.42 per cent.

H = 8.19 per cent., 8.27 per cent.

Acetyl (Freudenberg method) = 19.00 per cent.

Molecular weight (cryoscopic in phenol) = 207

(ebullioscopic in benzene) = 226

Rast method = $212, 217$.

$C_{13}H_{18}O_3$ requires C = 70.27 , H = 8.19

Molecular weight = 222 ; Acetyl = 19.4 per cent.

COLOUR REACTIONS OF CLERODIN

1. One milligram of *Clerodin* dissolved in 1 c.c. of glacial acetic acid is treated with 2 drops of ammonia. On warming the mixture an intensely deep pink colour slowly develops. Amyl alcohol extracts the colouring matter completely.

The amyl alcohol extract on evaporation leaves a residue which contains nitrogen in its composition. This colour reaction is very delicate and is characteristic of *Clerodin*.

The coloured solution gives a selective absorption band extending from 469μ to 507μ , as shown in the photo-reproduction (fig. 35). The colour reaction does not take place in neutral solvents instead of acetic acid.

2. *Liebermann's 'Cholestol' reaction*.—A small quantity of the substance was dissolved in acetic anhydride and a

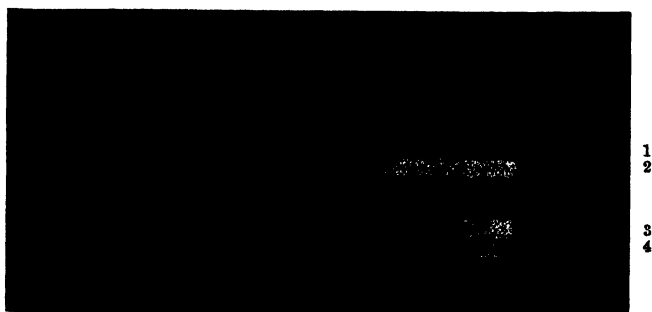


FIG. 35. 1, Copper arc spectrum ; 2 and 3, continuous ;
4, absorption spectrum of colour reaction product.

drop of concentrated sulphuric acid added, when a transient violet-pink coloration was produced. This changed to a beautiful green colour.

3. *Hager-Salkowski reaction*.¹—A small quantity of the substance was dissolved in glacial acetic acid (0.1 gram in 3 c.c.) to which 1 c.c. of chloroform was added and then a few drops of concentrated sulphuric acid. A yellowish-pink colour developed which changed rapidly to green.

DERIVATIVES OF CLERODIN

Acetyl Clerodin, $C_{13}H_{17}O_2.O.CO.CH_3$.—This was prepared by heating *Clerodin* (1.2 grams) with 15 c.c. acetyl chloride on a water bath for 1 hour. The excess of acetyl

¹ *Z. Physiol. Chem.*, vol. 57 (1908), p. 385.

chloride was evaporated off and the product was poured into water slowly with constant stirring, when a white flocculent precipitate was obtained. This was filtered, thoroughly washed with water and crystallised from dilute alcohol (85 per cent.) in short coarse rods melting at 110° C. It is lævo-rotatory, soluble in all ordinary solvents and is not bitter.

Found C = 68.00 per cent. ; H = 7.77 per cent.

Acetyl = 32.05.

$C_{13}H_{17}O_2.O.CO.CH_3$ requires C = 68.2 per cent. ;

H = 7.57 per cent.

Acetyl = 32.5.

Benzoyl Clerodin, $C_{13}H_{17}O_2.O.CO.C_6H_5$.—*Clerodin* (1.6 grams) was dissolved in pyridine (30 c.c.) and benzoyl chloride (10 c.c.) gradually added with vigorous shaking. The mixture was then heated on a water bath for half an hour, and poured into an excess of dilute hydrochloric acid containing lumps of ice. The benzoyl derivative was thrown down as an oily liquid. It was taken in ether and the resulting solution washed with dilute hydrochloric acid until free from pyridine. The ether extract was further washed with dilute sodium carbonate solution and finally with water. After complete removal of ether, a deep brown oil was obtained which did not solidify even after several months when kept in a vacuum desiccator over calcium chloride. The oil was then distilled at 140° C. under 1 mm. pressure. A clear and almost colourless oil was obtained, which on analysis gave the following results :

Found C = 73.21 per cent. ; H = 7.10 per cent.

$C_{13}H_{17}O_2.O.CO.C_6H_5$ requires C = 73.61 per cent. ;

H = 6.75 per cent.

Clerodin-phenylurethane, $C_{13}H_{17}O_2.O.NH.CO.C_6H_5$.—*Clerodin* (1 gram) was treated with phenyl isocyanate (5 c.c.) and the mixture warmed on a water bath for 1 hour. On allowing the product to stand at the room temperature for

one day, the phenyl-urethane derivative crystallised out. This was filtered off and washed repeatedly with petroleum ether and benzene until the smell of phenyl isocyanate had completely disappeared. The substance was then crystallised from alcohol in colourless silky needles melting at 240° C. with decomposition. The yield was extremely poor.

Found N = 4.4 per cent.

$C_{20}H_{28}O_4N$ requires N = 4.1 per cent.

Clerodin dibromide, $C_{13}H_{18}O_3Br_2$.—*Clerodin* (1.77 grams) dissolved in chloroform (50 c.c.) was treated with excess of bromine (4 c.c.) and the mixture allowed to stand for 4 hours in a dark place. The solvent and the excess of bromine were then distilled off over a water bath, when a pasty semi-solid mass was obtained. This on standing over quicklime in a desiccator for several days solidified into an orange-yellow vitreous mass. This on crystallisation from 90 per cent. alcohol (and charcoal) gave orange-coloured micro-crystalline powder, melting at 170° C. with decomposition.

Found Br = 41.6 per cent.

$C_{13}H_{18}O_3Br_2$ requires Br = 41.9 per cent.

Volumetric estimation of unsaturation.—*Clerodin* (0.2984 gram) was dissolved in carbon tetrachloride (10 c.c.) in a well-stoppered 250-c.c. bottle and 20 c.c. of N/3 bromine in carbon tetrachloride added. The mixture was allowed to stand in a dark place for 24 hours. The bottle with the mixture was then cooled in ice and 25 c.c. ice-cold water quickly added and the bottle well shaken. After this 25 c.c. of 10 per cent. aqueous potassium iodide solution and 75 c.c. water were again introduced, and the bottle thoroughly shaken. The free iodine thus liberated was titrated against N/10 thiosulphate. After the titration, 5 c.c. of 2 per cent. aqueous potassium iodate were added and the fresh iodine thus liberated again titrated. Twice this titration value was deducted from the first value, and

the equivalent of bromine atoms taken up by the *Clerodin* molecule calculated. This came to 2.01, which evidently indicates that the *Clerodin* molecule contains a double bond.

Mono-bromo-Clerodin-dibromide, $C_{13}H_{17}O_3Br.Br_2$.—The orange sticky mass left in the bottle after the foregoing titrations was recovered and after purification and recrystallisation from 90 per cent. alcohol (and charcoal) melted with decomposition at $110^{\circ}C$.

Found Br = 52.00 per cent.

$C_{13}H_{17}O_3Br.Br_2$ requires Br = 52.06 per cent.

IODINE VALUE OF CLERODIN

The iodine value of *Clerodin* was determined by Hanu's method. Complete absorption of iodine was observed after 2 hours' contact, when the value was found to be 115.00.

Iodine value calculated on the basis of one double bond came to 114.41.

By prolonging the time of contact erratic results were obtained.

ESTIMATION OF ACETYL GROUP IN CLERODIN

The estimation was effected by trans-esterification of 0.3434 gram of *Clerodin* in alcoholic solution by para-toluene sulphonic acid and titration, after hydrolysis, of the ethyl acetate removed by distillation.

Found acetyl = 19.00 per cent.

$C_{13}H_{18}O_3$ requires acetyl = 19.40 per cent.

Action of hydro-iodic acid on Clerodin.—*Clerodin* (0.367 gram) was heated with concentrated hydro-iodic acid (sp. gr. 1.7) in a current of dry carbon dioxide according to the classical method of Zeisel as described by Benedict and Bamberger¹ and modified by Gregor.² There was no evidence of the presence of any methoxy group.

¹ Benedict and Bamberger, *Monatsh*, vol. 11 (1890), p. 260.

² Gregor, *Monatsh*, vol. 19 (1898), p. 116.

It was, however, noticed that reddish-yellow flakes separated on cooling and standing overnight. These were filtered, washed free from hydro-iodic acid and after crystallisation from boiling acetone decomposed at 240° C. with previous softening at 220° C. The pure hydro-iodide, however, was not sufficient for a complete analysis.

Saponification value.—The saponification value of *Clerodin* was determined by heating a known weight of *Clerodin* with an excess of 0.1N alcoholic KOH under reflux for 1 hour and titrating back the excess of alkali. The completion of hydrolysis was accompanied with the formation of a crystalline precipitate. The saponification value was found to be 251 (theoretical value being 252). This figure was also obtained when *Clerodin* was hydrolysed by cold alcoholic potash, the time of contact being 4 hours only.

After the titration was complete the mixture was filtered from the crystalline precipitate and thoroughly washed with cold distilled water into a distillation flask. Excess of dilute sulphuric acid was then added to the filtrate and the mixture distilled in steam. The distillate was found to contain acetic acid only.

The crystalline precipitate obtained by the hydrolysis of *Clerodin* was recrystallised from boiling 90 per cent. alcohol in colourless prisms melting at 250° C. It is tasteless. It is a de-acetylated product of *Clerodin* and is a dihydroxy compound.

Found C = 73.01 per cent. ; H = 9.12 per cent.

$C_{11}H_{14}(OH)_2$ requires C = 73.33 per cent. ; H = 8.89 per cent.

The above compound was re-acetylated by heating with acetic anhydride in the usual manner. The acetyl derivative thus obtained was found after crystallisation from 90 per cent. alcohol to be identical in every respect with the mono-acetyl derivative of *Clerodin*. Melting point and mixed melting point of the two were found to be identical, 110° C.

The alcoholic mother liquor remaining after crystallisation of the acetylated product, when evaporated, left an extremely bitter substance possessing one acetyl group in the molecule. In appearance it seems to be identical with *Clerodin* itself. Further investigation on the subject is being continued.

SUMMARY

From the leaves of *Clerodendron infortunatum*, locally known as *Bhant*, a non-glucosidal bitter principle has been isolated in a chemically pure state. Several derivatives, for example acetyl, benzoyl, phenyl urethane, and bromo-compounds, have been prepared and a certain amount of light has been thrown on its chemical constitution. It appears to be an unsaturated hydroxy ester.

A specific colour reaction has been developed by which a minute quantity of *Clerodin* can be detected.

Its anthelmintic property has been verified by physiological experiments *in vitro*.

I take this opportunity of expressing my thanks to Sir J. C. Bose and Prof. N. C. Nag for the kind interest they have shown during the course of this investigation.

VII.—CHEMICAL EXAMINATION OF SEEDS OF *PACHYRHIZUS ANGULATUS*

BY

N. C. NAG, M.A., F.I.C., H. N. BANERJEE, M.Sc.,
AND A. K. PAIN, M.Sc.

PACHYRHIZUS ANGULATUS or *Dolichos bulbosus* is locally known as *Sank-alu*.¹ It is extensively cultivated in some parts of Bengal. The tubers, some of which weigh from 5 to 10 kilograms, are eaten raw. The tubers are coated with a thin fibrous skin, which can be easily peeled off. The fleshy portion inside the skin is juicy and sweet and perfectly white in colour. The following Table gives a statement of results of analysis :

TABLE I.—RESULTS OF ANALYSIS OF *SANK-ALU* TUBERS.
(Grown at Falta.)

	Per cent.
Moisture (at 100° C.)	80.0
Ash	1.1
Soluble reducing sugar	5.0
Carbohydrates transformable into reducing sugar by boiling with Hydrochloric Acid .	9.4
Proteins (Nitrogen 0.304 %)	1.9
Fibre	1.8
Total determined	<u>99.2</u>

Sank-alu tuber juice is rich in Vitamin C and has anti-scorbutic action on rats suffering from disease.

The subject of our main investigation is, however, the chemical examination of seeds of *Pachyrhizus* or *Sank-alu*.

¹ Nadkarni, *Indian Materia Medica*, pp. 312, 629; N. G. Mukherjee, *Indian Agriculture*.

The ripe seeds of *Sank-alu* are obtained from the dried mature pods, which yield pulse-like grains with a strong yellow colour. According to Nadkarni, the seeds are eaten either raw or cooked. In Bengal, however, they are not used for any other object than for sowing, there being a strong belief that the seeds prove to be poisonous when eaten by cattle or by human beings.

In view of the conflicting ideas as regards the edibility of the seeds, not only chemical but also physiological examinations of the seeds were undertaken. The results of the chemical analysis are given in Table II.

ANALYSIS OF THE SEEDS

TABLE II.—CONSTITUENTS OF *SANK-ALU* SEEDS

	Per cent.
Moisture (at 100° C.)	6.70
Oil (Ether extracted)	27.30
Proteins	26.25
Carbohydrates transformable into reducing sugar	20.00
Fibres	7.00
Ash	3.64
Total determined	<u>90.89</u>

The ash was next analysed, and the statement given below in Table III gives the different mineral constituents present in the ash.

TABLE III.—MINERAL CONSTITUENTS IN SEED ASH

	Per cent.
SiO ₂	1.09
Fe ₂ O ₃	0.68
P ₂ O ₅	33.01
CaO	13.00
MgO	6.96
MnO	0.05
Alkali Total K ₂ O + Na ₂ O	39.18
Total determined	<u>93.97</u>

The difference from 100 was made up of CO₂ and undetermined constituents such as Cl, SO₃.

EXAMINATION OF THE OIL

As the oil content of the seed was very high, a systematic analysis of the oil was carried out. The following Table IV gives the physical constants and Table V gives the chemical constants of the oil :

TABLE IV.—PHYSICAL CONSTANTS OF THE OIL

Yield	Specific gravity at 31° C.	Coefficient of expansion between 31° C. and 100° C.	Refractive index at 26° C.
27.3 %	0.9140	0.00049	1.4673

TABLE V.—CHEMICAL CONSTANTS OF THE OIL

Saponifica- tion value	Iodine value	Acid value	R.M. value	Insoluble fatty acid	Unsaponifi- able matter
196.7	85.3	1.1	2.71	93 %	2.35 %

The oil was saponified and the free acids liberated by the usual methods. After purification and dehydration the following constants were determined, which are given in Table VI :

TABLE VI.—CONSTANTS OF THE FREE FATTY ACIDS

Neutralisation value	Mean molecular weight	Iodine value	Melting point	Solidifying point
194.4	288	94.3	45° C.	38° C.

The free fatty acid mixture was then separated into the solid and liquid components by the Twitchell process. The results are given in Table VII.

TABLE VII.—CONSTANTS OF SOLID AND LIQUID ACIDS

Solid acid		Liquid acid	
Yield	Iodine value	Yield	Iodine value
37.6 %	7.99	62.4 %	132.8

It may be remarked in passing that the low iodine value of the solid acid is indicative of fairly good separation of the solid and liquid acids. Further, the presence of 37.6 per cent. of solid acid indicates its inferior quality as an edible oil.

UNSATURATED LIQUID ACID MIXTURE

The liquid acid mixture was then separated into its different components by the method of Eibner and Muggenthaler.

3.537 grams of oil were brominated in dry ether solution at -10°C . After bromination, no ether insoluble bromide was found to separate out, thus indicating the absence of high unsaturation and absence of linolenic acid.

Free bromine was then removed from the mixture by washing with aqueous thiosulphate, which latter was removed by further washing with cold distilled water. The ethereal solution was then evaporated to dryness and the mass dissolved in a minimum quantity of hot petroleum ether.

LINOLIC TETRABROMIDE

The petroleum ether solution on cooling deposited crystals of linolic tetrabromide. These were filtered and washed free from any dibromide by washing with small quantities of cold petroleum ether. The tetrabromide, after recrystallisation from boiling petroleum ether, melted

EXAMINATION OF SEEDS OF *PACHYRHIZUS ANGULATUS* 87
at 114° C. The crystals were thoroughly dried and weighed, from which the amount of the corresponding acid was calculated.

OLEIC DIBROMIDE

After removal of the tetrabromide, the petroleum ether solution was evaporated to dryness, when the oleic dibromide was left over as a brown oily matter. This was dried to constant weight, and from the weight thus obtained the quantity of oleic acid was calculated.

Statements of the results obtained are given in the following Tables VIII and IX :

TABLE VIII.—AMOUNTS OF BROMIDES OBTAINED

Oil taken	Ether insoluble bromide	Tetrabromide	Dibromide
3·537 grams	Nil	3·400 grams	3·056 grams

From the above results, the corresponding quantities of linolic and oleic acids were calculated, and hence their percentages, which are given in Table IX.

TABLE IX.—PROPORTION OF LINOLIC AND OLEIC ACIDS

Linolic acid calculated from tetrabromide	Percentage of linolic acid	Oleic acid calculated from dibromide	Percentage of oleic acid
1·587 grams	45 %	1·950 grams	55 %

From the above results the *Sank-alu* seeds appear to contain a high proportion of oil and protein matter. Hence it may be concluded that the seeds are likely to possess considerable nutritional value. This conclusion is further supported by the fact that high proportions of phosphoric acid, alkali and lime are present in the ash of the seeds.

PHYSIOLOGICAL EXPERIMENTS

In order to test the nutritive value of the seeds, experiments were carried out with rats, which did not show any willingness to take the crushed seeds as food. A mixture containing 75 per cent. B.D.H. diet and 25 per cent. crushed *Sank-alu* seed was then tried. The rats forced to take this mixed diet all died within a week, even before any pronounced symptoms of deficiency disease became evident. One of the symptoms exhibited was an inordinate thirst and gasping for breath.

The oil extracted from *Sank-alu* seeds was also found to be ineffective in curing disease in rats caused by deficient diet.

The next problem was to find out the particular constituent of the *Sank-alu* seeds which acted as the toxic agent.

ISOLATION OF A TOXIC SUBSTANCE IN THE SEEDS

For this purpose many experiments were carried out in regard to the action of various extracts on fishes. The following results were observed :

1. The effect of *Sank-alu* seed extract in water.—0.1 per cent. of seed powder in tap water killed fishes like *Tengra*, *Shing* and *Kai* within 30 minutes.

2. The effect of oil-free *Sank-alu* seed extract in water.—In this series of experiments the powdered seeds were thoroughly extracted with ether. The oil-free seed powder thus obtained was found to be even more effective in the sense that 0.1 per cent. of oil-free seed powder in tap water killed fishes in less than 15 minutes.

In order to find if it was possible to separate the toxic substance from the oil-free seed powder, the powder was treated with boiling alcohol. It was then found that the seed powder left over after a certain substance had been extracted from it by alcohol was no longer toxic. But the

alcoholic solution, after complete removal of the alcohol, gave a substance which was exceptionally toxic.

3. The effect of the alcoholic extract.—After thorough removal of the alcohol by evaporation, the residue was dissolved in water so as to produce solutions of three different strengths, 0·02 per cent., 0·01 per cent. and 0·002 per cent. Their effects on fishes were found as follows :

Strength of solution	Occurrence of death
0·02 %	After 17 minutes
0·01 %	After 18 minutes
0·002 %	After 60 minutes

NATURE OF THE TOXIC SUBSTANCE

From the above details it will be evident that there is a powerful toxic principle in *Sank-alu* seeds which is completely removed by boiling alcohol, leaving the seed powder perfectly harmless. The toxic substance is soluble in water, which answers to some of the characteristic tests for saponin.

SUMMARY

The seeds of *Pachyrhizus angulatus* (*Sank-alu*) have been chemically examined and the different constituents, such as mineral matter, oil content, protein content and carbohydrate content, determined. The oil has been analysed and determinations have been made of its different constituents. Feeding experiments have shown that the seeds of *Sank-alu* are unsuitable as food. They contain a toxic principle which answers to some of the characteristic tests for saponin.

We take this opportunity of expressing our grateful thanks to Sir J. C. Bose for his kind interest, helpful criticism and constant encouragement.

VIII.—NUTRITION EXPERIMENT WITH THE INDIAN FOODSTUFF *PISUM SATIVUM*

BY

N. C. NAG, M.A., F.I.C., AND A. K. PAIN, M.Sc.

In a previous paper¹ the value of *Cicer arietinum* as food has been discussed. It was there shown that *Cicer arietinum* can and does supply all the essential requirements for normal growth of rats from year to year, showing no sign of deterioration in health and in procreative function. The presence of oil soluble Vitamins A and D in *Cicer arietinum* was demonstrated by their curative effect on diseased rats. Various other pulses were also examined and were generally found to be deficient in food value.

In the present paper the relative food values of *Cicer arietinum* and *Pisum sativum* are the subjects of experimental investigation. The common field pea, *Pisum sativum*, locally known as *Mutur*, extensively cultivated in Bengal and Bihar during the cold season, is usually consumed in the green stage. The ripe seeds are consumed in various ways constituting the common *dal*. This *dal*, however, has not the same popularity as *Chhola* (*Cicer arietinum*) or as *Moong* (*Phaseolus radiatus*).

The procedure adopted in investigations on *Pisum sativum* is very similar to that described previously (*ibid.*).

EXPERIMENTS WITH *PISUM SATIVUM*

The following experiments were carried out with young rats born of a pair of healthy parents reared on *Cicer*

¹ Nag and Banerjee, 'On Oil Soluble Vitamins in *Cicer arietinum*,' *Trans. Bose Res. Inst.*, vol. ix, pp. 89-118.

arietinum diet. The litter under consideration was the third one of the parents, and consisted of 12 young ones, from which *one male and three females* were kept in Cage No. 1, while *one male and two females* were placed in Cage No. 2. These particular specimens were born on June 24, 1936, observations being commenced from August 3, after the weaning period.

The subjects in Cage No. 1 were then reared on wholemeal *Mutur* and water. The subjects in Cage No. 2 were placed at the same time on wholemeal *Kabuli chhola* diet. The rats were weighed every 3 days, and the results obtained are given in Table I. The experiments were all carried out in the year 1936.

TABLE I.—COMPARATIVE RESULTS OBTAINED AFTER FEEDING WITH *KABULI CHHOLA* AND *MUTUR*
(Vertical rows represent weights in grams)

Dates of observation	Aug. 3	Aug. 12	Aug. 29	Average increase in 26 days
<i>Mutur</i> Fed				
Male (a) . .	63	77	93	38 %
Female (b) . .	66	78	93	30 %
Female (c) . .	55	67	89	37 %
Female (d) . .	54	62	70	20 %
<i>Kabuli chhola</i> Fed				
Male (e) . .	51	70	107	70 %
Female (f) . .	57	71	99	54 %
Female (g) . .	56	69	93	50 %

Fig. 36 gives a graphic representation of the above results.

Analyses of the results detailed above show the difference in the gain of body weight under two different modes of feeding. Confining our attention first to the males, the *Kabuli chhola* fed male shows an increase of about 70 per

cent. in its weight, while the increase in the case of the *Mutur* fed male is no more than 38 per cent. In the case

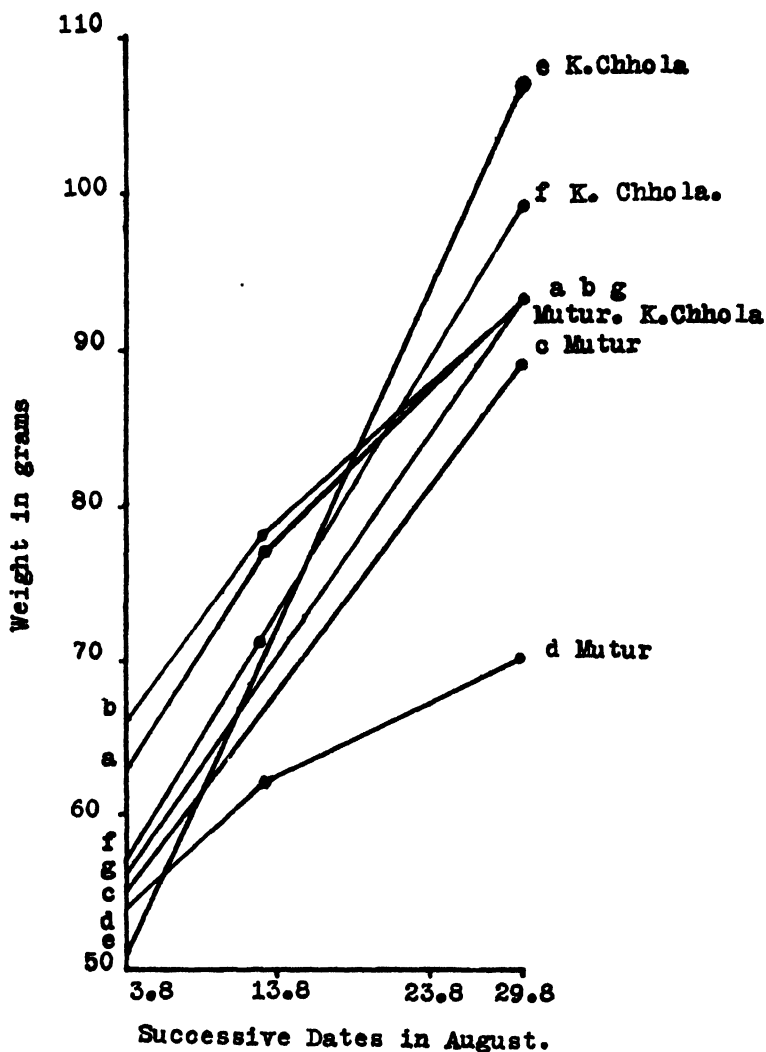


FIG. 36. Growth curves of rats, a, b, c, d (*Mutur* fed), and e, f, g (*Kabuli chhola* fed).

(In the above a and e are males.)

of the females also there is a considerable difference in the gain of weight in the two experimental batches ; those fed with *Kabuli chhola* gained about 50 per cent., while those fed with *Mutur* gained only about 30 per cent.

The males, generally speaking, gain relatively more in weight than the females.

In general appearance, *Kabuli chhola* fed rats appeared to be bigger and stronger. There was, however, no actual outward indication of deficiency disease in the *Mutur* fed batch.

The two batches of rats, born on June 24, have been kept under observation for a considerable length of time on their respective diets. A certain fact was noticeable in the meanwhile. The female (*f*), fed on *chhola*, gave birth to its first litter of 3 young ones on October 4, 1936, and the female (*g*), also fed on *chhola*, gave birth to its first litter of 7 young ones on October 9, 1936, only 5 days later. This confirms our observation described in our previous paper that in *chhola* fed rats the age when the first litter appears is generally between 3 and 4 months. In the present case the birth of the litters occurred in the course of about 3·5 months.

But the *Mutur* fed set of three females and one male has, even after the lengthened period of 8 months, failed to give a single litter. As the power of procreation is supposed to be due to the presence of Vitamin E in the food, the absence of any litter in the *Mutur* fed batch may perhaps be due to the deficiency of this element in the food.

FURTHER EXPERIMENTS WITH *PISUM SATIVUM*

The procedure adopted in the following set of observations was slightly different. The subjects for the experiments were chosen from two litters, 16 young ones in all, born on the same date, July 30, 1936. The parent animals, one male and two females, had all been normally fed on *chhola* diet.

Out of these 16 young ones, four males and four females were chosen and placed in four cages, numbered (11), (12), (13) and (14), one male and one female being in each cage.

The cages numbered (11) and (12) were supplied with devitaminised B.D.H. food from September 2, 1936, when observations were started after the weaning period of about a month. The period of devitaminisation lasted till September 23, 1936, *i.e.* for 3 weeks. After this period Cage No. (11) was supplied with *Kabuli chhola* diet, while Cage No. (12) was supplied with *Mutur* diet. The effects observed are given in Table II with explanatory details.

TABLE II.—EXPERIMENTS ON RATS FED ON DEVITAMINISED DIET
(Vertical rows represent weights in grams)

Dates of observation	Sept. 2	Sept. 9	Sept. 16	Sept. 20	Remarks
B.D.H. Food					
Cage (11) Male .	30·0	38·0	41·0	41·0	Eyes affected
Cage (11) Female .	36·0	45·0	47·5	47·0	Eyes affected
Cage (12) Male .	44·0	49·0	52·0	58·5	
Cage (12) Female .	40·0	44·5	48·5	49·5	

As will be seen from the above, both the male and the female in Cage No. (11) suffered from deficient food, the eyes becoming affected. But the male and the female in Cage No. (12) were not yet affected. The difference is due to the fact that both the male and the female in Cage No. (12) were, to start with, heavier in weight and more vigorous.

The B.D.H. diet of Cage No. (12) was continued till September 26, by which time the subjects showed stoppage in the gain of weight after having attained the weights of 62·5 grams (male) and 50·5 grams (female). When the gain

in weight was stopped under B.D.H. diet, these devitaminised rats in Cage No. (12) were henceforth fed on *Mutur* diet from September 26. This resulted in a continuous gain in weight in the male from 62.5 grams on September 26 to 81 grams on October 3 and to 97 grams on October 14. In the case of the female rat in Cage No. (12), the weight increased continuously from 50.5 grams on September 26 to 69 grams on October 3 and to 90 grams on October 14. The devitaminised rats thus regained their growth under *Mutur* diet and there was no outward sign of any deficiency. The fact that feeding with *Mutur* impaired the power of procreation became, however, evident from the fact that even as late as March 1937 (*i.e.* when the rats were 8 months old) the particular pair of rats did not give birth to any litter.

In Cage No. (11) the B.D.H. deficient diet had stopped gain in weight by September 20, and produced deficiency disease, evidenced by the eyes becoming affected. After 3 days *Kabuli chhola* diet was commenced on September 23. The change of diet was at once followed by continuous gain in weight, which in the male increased from 41 grams on September 23 to 43.5 grams on September 26, to 61 grams on October 3 and to 84 grams on October 14. The weight of the female, which had fallen to 45.5 grams on September 23, increased to 54 grams on September 26, to 76 grams on October 3 and to 97 grams on October 14. The eye disease was moreover completely cured. Further, the pair has been giving litters at regular intervals.

It is to be noted in this connection that after the supply of *chhola* diet, the deficiency disease of both the male and the female was completely cured. Far more important is the fact that after the supply of *chhola*, the female in Cage No. (11) gave birth to healthy litters at regular intervals, the young ones being as healthy and vigorous as the parents. In contrast with the failure of procreation in the *Mutur* fed batch, supply of *Kabuli chhola* restored the power of procreation.

SUMMARY

Rats grow normally when fed on *Cicer arietinum* (*Kabuli chhola*). *Cicer arietinum* acts as a curative for deficiency disease brought on by devitaminised food. *Pisum sativum* (*Mutur*) in the ripened state seems to serve as diet without bringing on any disease in rats, at least outwardly, though the gain in weight of the subjects is generally below normal. Rats, when very slightly affected by devitaminised food, seem to recover when placed on *Mutur* diet. It has, however, been found that rats fed on *Mutur* diet are unable to discharge the function of procreation. It would therefore seem that there is a certain vitamin deficiency in *Mutur* or that there is something in *Mutur* which inhibits or destroys the power of procreation.

We take this opportunity of expressing our grateful thanks to Sir J. C. Bose for his kind interest, helpful criticism and constant encouragement.

IX.—INTERSPECIFIC HYBRIDISATION IN *ORYZA*

I. CYTOGENETICAL EVIDENCE OF THE HYBRID ORIGIN OF *ORYZA MINUTA* PRESL

BY

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F.R.M.S.

INTRODUCTION

ORYZA MINUTA Presl, a distinct species of rice, had been variously classified as *O. officinalis* Wall., *O. latifolia* Desv., and *O. manilensis* Murr., by the taxonomists. From an analysis of morphological characters it is evident that *O. minuta* is very closely related to *O. officinalis* Wall. and *O. sativa* L. It is, however, smaller in all parts than either of the other species and represents a combination of the characters which distinguish *O. sativa* and *O. officinalis* from each other. The high fertility and true breeding habit of *O. minuta* seem inconsistent with its hybrid nature.

In a recent paper (Nandi, 1936 B) I have shown that the taxonomical separation of the species of *Oryza* is in association with a cytological separation into two groups, viz. (1) one group with $n = 12$ and (2) the other group with $n = 24$. From a preliminary cytological study of this species it was suggested that *O. minuta* had arisen from the hybridisation of *O. officinalis* \times *O. sativa*, followed by a chromosome doubling in the F_1 hybrid.

In its high fertility, constancy and formation of bivalents, *O. minuta* is analogous to other allopolyploids.

Within recent years various workers have discovered autotriploid and autotetraploid races of *O. sativa* in the common rice fields (Nandi, 1936 A). But polyploidy within the species of *Oryza* was unknown until the author (Nandi, 1936 B) first discovered it. I have already shown (Nandi, 1936 B) that *O. sativa* itself is not a true diploid species but a secondary balanced allotetraploid having $n = 12$ as the present existing haploid number (a secondary number derived from a basic number of 5).

Most of the species of *Oryza* are characterised by the number $n = 12$ and polyploidy within the species runs in multiples of 12. The question naturally arose whether *O. minuta* is an auto-octoploid or allo-octoploid. A comparative study of the morphology of chromosomes of *O. sativa*, *O. officinalis* and *O. minuta* and chromosome affinities, whether autosyndetic or allosyndetic, in the meiosis of the F_1 hybrids among these species was therefore undertaken to elucidate the genomic constitution of *O. minuta*.

The results so far obtained show that *O. minuta* represents a clear verification of Winge's theory (1917) that multiple chromosome number among polyploid is the result of species hybridisation followed by chromosome doubling.

Further results will be described later, on interspecific hybridisation between different species of *Oryza*.

MATERIAL AND METHODS

The investigations were carried out in the greenhouse at the Genetical Laboratory, Regent's Park, London (1934-35), and in the Bose Research Institute, Calcutta (1936-37).

Several crosses were made between *O. sativa* and *O. officinalis*, using *sativa* as the female parent. Of these crosses only two produced ripe seeds. Crosses were also made in the opposite way with *officinalis* as the female parent; in this latter case it was found that the flowers dropped away before ripening of the seeds.

In the next year two plants were raised from the ripe seeds as obtained above. Phenotypically the hybrid plants looked more like *sativa*. The anthers were considerably smaller in size and nondehiscent, containing only shrivelled pollen grains. The stigma of the two specimens was pollinated by *sativa* and *officinalis* pollen without success. The plant was highly sterile and did not produce any seed when thus pollinated.

In the year 1935, hybridisation experiments were carried out between *O. minuta* \times *O. officinalis* and *O. minuta* \times *O. sativa*, using *minuta* as female in both cases. The resulting hybrids obtained in the next year were found to be sterile.

Root-tip materials were obtained by germinating seeds on moist filter papers in Petri dishes. Germination was rapid and the tips were ready in four days. Fixations were made with Navashin, 2BE (La Cour, 1931), and a modification of Benda. Of these three fixatives the last was found to be very successful, its formula being :

1 % Chromic acid	20 c.c.
2 % Osmic acid	4 c.c.
Glacial acetic acid	1 c.c.
Maltose	0.4 grm.

Somatic metaphase plates were well spaced out. The constrictions of the chromosomes were very clear, so that a detailed study of their morphology was possible.

Studies on the meiosis were made from mounts in Belling's iron aceto-carmin fluid. The buds were collected between 1 and 3 P.M. on sunny days. The glumes of the buds were cut off at the tips and fixed in Navashin's fluid for 4 hours. An exhaust pump was always used. Penetration was very rapid and fixation perfect.

All the paraffin materials were cut at 14 μ in thickness and stained with iodine-gentian-violet. The slides were mordanted in $\frac{1}{2}$ per cent. chromic acid for 20 minutes and rinsed in water before staining ; this gave excellent results.

Owing to the limited number of the seeds the root-tips of the hybrids could not be studied.

SOMATIC CHROMOSOMES

The details of somatic chromosomes of *O. sativa* have already been described (cf. text fig. 1, Nandi, 1936 B, and also Plate I, 1). Examination of the somatic chromosomes of *O. officinalis* reveals that the chromosomes are definitely bigger in size than those of *O. sativa* (Plate I, 2). Analysis of the morphology of the chromosomes shows the same types of chromosomes as reported in *O. sativa*. Of the 10 types of chromosomes present, eight are present twice and two are present four times, making a total of $2n = 24$, in the apparently diploid complex of *O. officinalis*.

The chromosomes of *O. minuta* show two classes of different sizes. In the first the 24 chromosomes were small and in the second the other 24 chromosomes were relatively large (Plate I, 3). Several drawings of somatic plates were made for the detection of different types of chromosomes and their classification by their size and attachment-constrictions. The result of this study is depicted semi-diagrammatically in Plate I, 4, which represents the combined basic sub-genome of *O. minuta*. It is clear that there are present 10 types of chromosomes of two quite different sizes. Of each of these two 10 types of chromosomes, eight are present twice and two types are present four times in the haploid genome of *O. minuta* ($n = 24$).

The members of these two size groups are designated $s_1 \dots s_{10}$ and $o_1 \dots o_{10}$ respectively, s representing the chromosomes of *sativa* and o those of *officinalis*. The size of the chromosomes and the number of satellites suggest the possibility that the chromosomes of *O. minuta* consist of two distinct sets from two different species, viz. one group of 24 small chromosomes presumably belonging to *O. sativa*, and the other group of 24 large chromosomes belonging to

O. officinalis. That is, the chromosome complements of *O. minuta* do not represent a reduplication of a basic set. The evidence from somatic chromosomes alone seems to indicate that *O. minuta* originated by hybridisation of two different non-homologous chromosome sets followed by a doubling of the chromosome numbers.

MEIOSIS

Studies on the meiosis of *O. sativa* and *O. officinalis* have been previously reported (Nandi, 1936 A, B). A comparison of the meiotic chromosomes of the two species at metaphase I shows a distinct size difference (Plate I, 5, 6). It is, however, evident that there is no difference in chromosome configurations, the chiasma frequency being the same in both the species.

A variable amount of secondary association among the bivalents at metaphase I or the univalents at metaphase II is also found in *O. officinalis*. The maximum association observed is two groups of three bivalents and three groups of two bivalents at metaphase I (Plate I, 7). It is therefore consistent with the view already expressed (Nandi, 1936 B) that the existing species of *Oryza* with an apparent haploid number of 12 are secondarily balanced, the basic chromosome number being 5.

Anaphase and second division proceed regularly, followed by the formation of normal pollen grains.

Certain aberrations in the meiotic divisions of *O. officinalis* were noticed which call for special attention.

Diploid pollen grains were observed in some of the plants. The processes of the formation of polyploid gametes have been studied in detail in *O. sativa* (Nandi, 1936 A). Plate I, 8, shows a cell with 24 bivalents at diakinesis instead of 12 bivalents, the size of the cell being twice the size of the normal cells. This giant cell has probably arisen owing to the fact that the two nuclei formed by the nuclear division

of the archesporial cell were located near each other and in each of them synapsis proceeded independently, after which the disappearance of the nuclear membrane took place. Absence of quadrivalents to be expected in such an auto-tetraploid nucleus shows that nuclear union here had been prior to synapsis. The formation of diploid pollen grains is expected by a normal behaviour of the chromosomes in this cell. In Plate I, 9, there are 24 univalents instead of 12 bivalents. This lack of pairing among the chromosomes at metaphase I is either due to failure of synapsis and consequently failure to form chiasmata, or due to premature separation of the chromosomes after normal pairing. Owing to the failure of the development of the spindle in the nucleus the univalents fail consequently to be distributed between the two poles leading to the formation of a 'restitution nucleus' (Rosenberg, 1927). The nucleus is then turned into second division stage, where the univalents split lengthwise in the same manner as in normal metaphase II, but with the diploid number of chromosomes. These chromosomes now separate between the poles and form two nuclei with 24 chromosomes in each. The dyads formed in this way develop directly into two diploid pollen grains with double the number of chromosomes.

MEIOSIS OF THE HYBRID

O. sativa × *O. officinalis*

At the stage corresponding to diakinesis, 24 univalents were observed (Plate I, 10). Here it is seen that there are distinct size differences between the 24 chromosomes as observed in *O. sativa* and *O. officinalis*. The fact that interspecific hybrids of *Oryza* show absence of bivalents leads one to conclude that the chromosomes of the respective parent species are too well differentiated for a regular meiotic pairing. The application of X-rays to the chromosomes of *Drosophila*, *Datura*, *Crepis*, etc., has shown that

the formation of new species is preceded by some types of chromosome rearrangements resulting in a certain degree of cross sterility, which sets up a physiological isolation within the parent species. Studies on the morphology of the chromosomes of the two species of *Oryza* have shown that such changes in chromosome size and diameter are external expressions of corresponding differences due to genic and structural changes in the chromosomes themselves. It seems, therefore, logical to assume that changes in chromosome affinity between the species will likewise change and would manifest itself by the almost complete absence of synapsis.

As a result of these chromosome transformations and altered affinity, bivalents are not formed and univalents are found scattered throughout the spindle (Plate I, 11); consequently, meiosis does not proceed normally and the irregularities in Divisions I and II result in degenerating microspores of varying size and number. The pollen grains are found empty and devoid of starch—a result to be expected in view of the aberrant meiosis.

Meiosis of O. minuta

A study of the meiosis of *O. minuta* had already been made (Nandi, 1936 B). At metaphase I, 24 bivalents of two quite distinct size-classes are formed, no multivalents or univalents being observed. Secondary association between the two sets of markedly different sized chromosomes, belonging to the two parental species, is clearly evident at metaphases I and II (Plate I, 12, 13). Here the maximum association is 12 groups of two bivalents or univalents at metaphase I or II respectively. That is, the basic number of *O. minuta* is 12. Absence of quadrivalents and presence of bivalents and secondary association indicate that the chromosomes of *O. minuta* do not represent a reduplication of a single set but are derived from two different species with genetically dissimilar chromosomes.

Both anaphase and second meiotic division in this species proceed normally. Pollen grains were examined in a drop of lactophenol containing cotton blue. They proved to be viable in all cases.

Meiosis of the Hybrids

O. minuta ($n = 24$) \times *O. officinalis* ($n = 12$)
and

O. minuta ($n = 24$) \times *O. sativa* ($n = 12$)

In the hybrids, meiosis was found to be very irregular. At metaphase I, 12 bivalents and 12 univalents were generally present; trivalents and quadrivalents were not found at all.

In the cross *O. minuta* \times *O. officinalis*, with 36 chromosomes (Plate I, 14), it is found that 12 bivalents are bigger, the two chromosomes of each bivalent being of the same size and shape. This indicates that pairing of the homologues is autosyndetic, and consequently the 24 chromosomes, consisting of 12 bivalents, belong to a similar set, *i.e.* 12 chromosomes of *O. officinalis* have found 12 homologues in the haploid genome of *O. minuta* and have formed 12 bivalents. The rest of the 12 univalents are smaller in size and belong to another set.

Again, in the hybrid *O. minuta* \times *O. sativa*, with 36 chromosomes (Plate I, 15), it is seen that 12 bivalents are smaller, the two homologous chromosomes of each bivalent being of similar size and shape. This shows that pairing is autosyndetic, and consequently these 24 chromosomes, consisting of 12 bivalents, belong to another similar set, *i.e.* 12 chromosomes of *O. sativa* have found 12 homologous chromosomes in the haploid genome of *O. minuta* and have formed 12 smaller bivalents. The other 12 univalents are bigger in size than the bivalents and consequently belong to a different set. It is also found that conjugation of the chromosomes in either hybrid never occurs between small and large ones but always between small ones or large ones.

That the hybrids with 36 chromosomes do not contain one similar set of chromosomes is evident from the absence of trivalents or multivalents.

A comparison between metaphase I chromosomes in *O. sativa* and *O. officinalis* and the two hybrids seems to show that the difference in chromosome size as observed in *O. sativa* and *O. officinalis* corresponds to the differences in chromosome size observed in the two sets of chromosomes in the hybrids. Evidently, in *O. minuta*, with 24 bivalents, one set of 12 bigger bivalents is derived from *O. officinalis* and the other set of 12 smaller bivalents is derived from *O. sativa*.

The chiasma frequencies have been determined from the pure species and the hybrids (Table I).

TABLE I.—THE AVERAGE CHIASMA FREQUENCY PER BIVALENT

Species	Stage	Number of nuclei	Number of pairs of bivalents	Mean number of chiasmata per bivalent
<i>O. sativa</i>	Metaphase	15	180	1.5
<i>O. officinalis</i>	"	15	180	1.5
<i>O. minuta</i>	"	15	360	1.5
<i>O. minuta</i>	" .	15	180	1.5
<i>O. officinalis</i>				
<i>O. minuta</i>	"	15	180	1.5
<i>O. sativa</i>				

It is evident that there is no significant difference in chiasma frequencies between the hybrids and the pure species.

Anaphase and second divisions in the hybrids show a number of irregularities. As a consequence of such irregular divisions a large proportion of the pollen grains were empty and the variation in their size well marked.

DISCUSSION

The phenomenon of polyploidy, *i.e.* the duplication of the chromosome complex, has been subject to intensive research. This condition is of very frequent occurrence in the plant kingdom, but not so common in animals. The problem of the multiple chromosome series became still more important when different workers succeeded in producing polyploid plants experimentally. Müntzing (1932) re-created an existing Linnean species of *Galeopsis Tetrahit* ($n = 16$) by the hybridisation of two other species, *G. pubescens* ($n = 8$) and *G. speciosa* ($n = 8$), with lower chromosome numbers. Then the question arose whether the natural polyploids have arisen in a similar way to those of the polyploids produced experimentally.

It was soon realised that polyploidy was of two kinds. The terms autopolyploidy and allopolyploidy were first introduced by Kihara and Ono (1927), the former denoting polyploidy resulting from the reduplication of similar sets of chromosomes from the same species and the latter from the reduplication of different sets of chromosomes derived from the hybridisation of different species.

An autotetraploid plant was first discovered by de Vries (1913) in his *Oenothera* cultures. This tetraploid mutant, *Oenothera Lamarckiana gigas*, originated through a longitudinal split in the chromosomes in the fertilised egg (Gates, 1909). Various autopolyploid races of plants have since been discovered or experimentally produced.

In a recent paper (Nandi, 1936 A) it has been shown that diploid gametes are of frequent occurrence in the varieties of rice. All the methods for the formation of unreduced gametes in the different species of plants so far reported are observed in *Oryza sativa* and *O. officinalis*, viz. (1) complete suppression of the first division as a result of failure of pairing; (2) pollen mother-cell fusion; (3) syndiploidy; (4) failure of separation of the univalents in the second

division ; (5) failure of cytokinesis in both the divisions, all of which lead to the formation of diploid gametes. They have also been produced experimentally by raising the temperature of the greenhouse (Nandi, 1936 c). The frequent occurrences of autotriploid and autotetraploid races of rice plants have been shown to be due to the function of these diploid gametes.

If diploid gametes can arise in the laboratory through physiological disturbances or external factors, there is no reason why they should not arise also in nature under extreme conditions of heat and cold. And there is no reason to doubt that most of the natural polyploid plants have originated in this way.

Opinions differ as to the evolutionary importance of autopolyploidy. It is generally found that autopolyploidy leads to increased size of nuclei and cells and consequently effects a greater increase in the size of all parts of plants. Autopolyploid races of plants in contrast with allopolyploids are characterised by multivalents and are unstable and revert to the original diploid type in succeeding generations. They can survive only by means of apomixis and vegetative propagation.

On account of the rarity of autopolyploid species of plants in nature and the difficulty of explaining the mechanism by which quadrivalent formation leads to bivalent formation, the writer in agreement with v. Wettstein (1927, 1928, 1932) and Clausen (1926) considers that autopolyploidy is of no evolutionary significance in the origin of species.

On the other hand, the second method of duplication of chromosomes, viz. allopolyploidy, has played an important rôle in the formation and evolution of species. This type of duplication arises from the hybridisation of two different species and by subsequent chromosome doubling in the sterile hybrid either somatically or gametically. *Primula kewensis* ($n = 18$) is a classical example which originated

from a cross between *P. floribunda* ($n = 9$) and *P. verticillata* ($n = 9$) in which a branch arose with twice the number of chromosomes due to a suspended cell-division in the F_1 hybrid (Newton and Pellew, 1929).

Numerous cases of experimental production of new species by wide species crossing or generic hybridisation with unreduced gametes are well known in recent years. Rybin (1929) produced a new *Nicotiana Tabacum-sylvestris* species by chance union of two diploid gametes formed in the sterile F_1 hybrid between *N. Tabacum* and *N. sylvestris*. This tobacco species created experimentally has maintained itself through successive generations. Karpechenko (1927), in a cross between diploid *Raphanus sativus* and *Brassica oleracea*, obtained a sterile hybrid which produced diploid gametes and gave origin to a new tetraploid genus, *Raphano-Brassica*, which is fully fertile and constant.

In the genus *Oryza* it is found that most of the species are characterised by the number $n = 12$, the present existing haploid number. *O. minuta* ($n = 24$) is the only known species with double the number of chromosomes.

A study of the morphology of chromosomes of a few species in a genus and chromosome conjugation in the hybrids, either autosyndetic or allosyndetic, indicates the methods of origin of polyploid species with lower chromosome numbers.

The question naturally arose, by what processes the duplication of the chromosome number in the species *O. minuta* was affected; whether by autopolyploidy or by allopolyploidy?

Thus a comparison of the morphology of chromosomes between two species of *Oryza* and breeding experiments show that species differentiation with the same chromosome number has taken place by gene mutations and structural changes in the morphology of the chromosomes, such as inversions, translocations, reduplications, deletions, unions and fragmentations.

Similarly, from a study of both somatic and meiotic chromosomes it is quite evident that the chromosomes of *O. minuta* may be divided into two groups on the basis of chromosome sizes. Skovsted (1934) has shown that New World Cottons ($n = 26$) are allopolyploid, originated from the doubling of chromosomes in a hybrid between two other species of *Gossypium* ($n = 13$) in each, the chromosomes of the parents being recognised by the differences in the morphology of the chromosomes.

Hybridisation experiments between *O. sativa* and *O. officinalis* show complete lack of synapsis and the differences in the chromosome sizes are maintained in the hybrid. It is therefore reasonable to explain this non-homology of the chromosomes in the sterile hybrid as due to structural and genetic changes in the chromosomes and is an indication that such alterations subsequently have become sufficiently distinct from the original ancestral species to be ranked as new species.

From a study of somatic chromosome analysis, meiotic chromosome association, complicated genetic results and the genus nearly related to *Oryza*, it has been shown (Nandi, 1936 B) that the genus *Oryza* originated through hybridisation between two different five-paired species in which two chromosomes were duplicated. This, followed by a subsequent doubling of the chromosomes, attained the secondary balance of $n = 12$, the present fundamental number for the existing species of *Oryza*. Haploid derivatives of *O. sativa* do not seem to contain any homologous chromosomes, *i.e.* the two sets of chromosomes are sufficiently distinct and do not represent a reduplication of a basic set.

If transformations of the chromosomes in the differentiation of the species of *Oryza* occur after polyploidy, as shown by the differences between *O. sativa* and *O. officinalis*, it is reasonable to assume that they must have also occurred before polyploidy arose.

In the hybrid *O. minuta* ($n = 24$) \times *O. officinalis* ($n = 12$).

12 larger bivalents are formed, while the other 12 chromosomes usually remain as univalents. Similarly, in the hybrid *O. minuta* \times *O. sativa* 12 bivalents are formed which are smaller in size than the rest of the 12 univalents. Here both the hybrids have approximately the same chiasma frequency. It is also generally found that 12 is the minimum number of univalents in each hybrid. This shows that in the hybrids with 36 chromosomes, 24 chromosomes are homologous, having the same pairing blocks which result in 12 bivalents (autosynopsis), and the rest of the 12 chromosomes are non-homologous and remain as univalents. Again we find that *O. minuta* is characterised by 24 bivalents, and on the basis of chromosome morphology, the two sets of 12 bivalents can be recognised by their markedly different size. Further, the chiasma frequency in the pure species of *O. sativa*, *O. officinalis* and *O. minuta* and the two hybrids is the same, there being no significant difference in the hybrids with 12 bivalents.

Evidently *O. minuta* is an allopolyploid (modified octoploid) formed by natural hybridisation between *O. officinalis* and *O. sativa* with double the number of non-homologous chromosomes; and the two species overlapping in geographical distribution permitted such hybridisation. In such a species there would be bivalents (autosynopsis), good fertility and evidence of secondary association due to remote affinities between the parental sets of chromosomes of common ancestry. Quadrivalent formation would never occur in this allopolyploid as the chromosome sets are too well differentiated to permit of multivalent chromosome associations.

Considering the widespread occurrence of diploid gametes in *O. sativa* and *O. officinalis*, it is quite probable that *O. minuta* arose from the fertilisation of a diploid egg of *O. officinalis* by a diploid pollen grain from *O. sativa*. Otherwise, if the chromosomes of *O. minuta* were derived from the multiplication of a basic set of 12, we should have

encountered four chromosomes of each type represented four times in the set and it would have been characterised by the formation of quadrivalents at meiosis. *O. minuta* is therefore not an autopolyploid.

There is thus good evidence that new species can arise from the already existing species by hybridisation as the result of diploid gametes formation. This example thus vindicates Winge's theory of the formation of polyploid species by hybridisation, which is an important factor in evolution (Winge, 1917, 1932). The demonstration that diploid gametes formation occurs frequently, leading to the production of allopolyploids, shows that duplication of the entire chromosome sets in the hybrids is of great importance for the economic development of interspecific hybrids and synthesis of new species of plants.

The present study shows that there are two different sets of chromosomes in *O. minuta*, one set of 24 from *O. officinalis* and the other set of 24 from *O. sativa*.

Autosyndesis with 24 bivalents in *O. minuta* is the general rule, i.e. o_1^* pairs with o_1 and s_1^* pairs with s_1 . During segregation of these bivalents at the reduction division the gametes of *O. minuta* therefore consist of two sub-genomes, each gamete being of the composition $o_1s_1 \dots o_{12}s_{12}$. Such formation of gametes gives on fertilisation a zygote of the constitution $o_1o_1s_1s_1 \dots o_{12}o_{12}s_{12}s_{12}$, and consequently this leads the species to breed true, restore fertility and remain constant.

Cytogenetical studies have shown that different degrees of homology exist between the chromosome sets of polyploids, and in the absence of homologues, chromosomes that might be considered to be non-homologous are found to pair.

The hybrid *Crepis setosa* \times *C. biennis* (Collins and Mann, 1923) is often cited as one of the best known examples of autosyndesis. *Crepis biennis* is regarded as an autopolyploid

* $o_1o_1 \dots o_{12}o_{12}$ represent 12 bigger bivalents in *O. minuta* presumably from *O. officinalis*, and $s_1s_1 \dots s_{12}s_{12}$ represent 12 smaller bivalents presumably from *O. sativa*.

species by Darlington (1932). Recently, Babcock and Swezy (1935) from a careful study have shown that in *C. biennis* there are 10 types of chromosomes which are probably present four times, making the present number $2n = 40$. The 10 types of chromosomes fall into 5 groups, each with two rather similar chromosomes. This analysis of chromosome types has led the authors to conclude that *C. biennis* has originated by hybridisation between two 5-paired closely related species followed by a doubling, thus producing a 10-paired allopolyploid species, and this succeeded by a second doubling gave rise to the present 20-paired species of *C. biennis*. The occurrence of bivalents in *setosa* \times *biennis* is no doubt due to autosyndesis, there being two genomes of one species and two of the other species concerned in the origin of *setosa* forming 10 bivalents. In a 'back cross' (Collins, Hollingshead and Avery, 1929), *C. artificialis* (10 *biennis* + 2 *setosa*), the 10 *biennis* chromosomes form 5 bivalents, no doubt due to allosyndesis. This is consistent if the *biennis* originated, as suggested by Babcock and Swezy (1935), as a hybrid between two closely related species or sub-species. Similarly it is known that in *Primula floribunda* \times *P. verticillata* (Newton and Pellew, 1929) the chromosomes from the two species regularly pair in the absence of competition, whereas in the tetraploid *P. kewensis* the chromosomes of the parental sets pair among themselves.

If this is the case, *Crepis biennis* should be considered as an allo-octoploid species and not an autopolyploid, as Darlington (1932) assumed. This shows that one must be guarded in drawing conclusions from the chromosome conjugation alone in a hybrid, whether as the result of autosyndesis or allosyndesis. Sharp (1934) points out that 'When the origin of the heteroploid condition is not directly known, an examination of the morphology of the chromosomes composing the complement often yields decisive evidence as to the kind of heteroploidy represented. Such

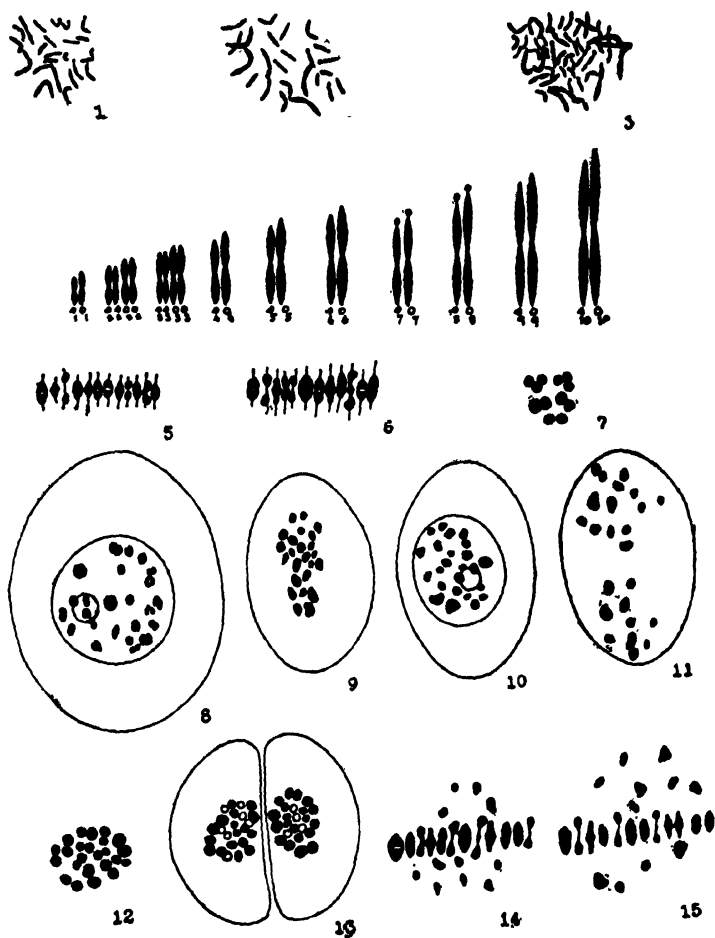
studies afford invaluable evidence supplementing that of the geneticist, taxonomist and student of phylogeny.'

Studies of Babcock and Swezy (1935) and Nandi (1936 B) will no doubt throw further light on the uncertainty of a few more of the autopolyploid species. It has already been pointed out by Winkler (1916) that the tetraploid tomatoes and *Solanum nigrum* are not to be considered as new species, but are tetraploid races of plants.

It is therefore reasonable to conclude that autopolyploidy is of little importance in the evolution of plant species. On the other hand, allopolyploidy must have played a significant rôle in evolution and species formation.

In comparing the morphology of the three species of *Oryza* it is found that *O. minuta* is intermediate in character. It is generally known that allotetraploid plants are usually more vigorous with large leaves, flowers and seeds, and cell-size is greater than in diploids. Thus there is a positive correlation between chromosome numbers and 'gigas' characters, no doubt due to heterozygosity. It is, however, found that there is an optimum for such correlation beyond which polyploid species become less vigorous. Stählin (1929) finds that vigour generally increases from diploidy to hexaploidy in *Poa* and *Festuca*. Octoploid species in these genera are found to be smaller in size than tetraploid and hexaploid species. Hexaploidy therefore seems to represent the optimum increase of cell-size in these two genera. Similarly the apparently diploid species of *Oryza* which are secondarily balanced allotetraploids seem to represent the optimum increase in vigour. As compared with the other species of *Oryza*, *O. minuta* (a modified octoploid) species is much smaller in size of all the parts and corresponds to the octoploid species of *Poa* and *Festuca*.

Hexaploidy, however, seems to be absent in *Oryza*. Thus, although the production of hexaploid species of *Oryza* from a cross between *O. minuta* and *O. sativa*, with subsequent doubling of the chromosomes, is theoretically possible, it is

FIG. 37. Nandi on *Oryza*.

EXPLANATION OF PLATE I

1. *O. sativa* ($2n = 24$). Somatic chromosomes at metaphase. ($\times 3200$.)
2. *O. officinalis* ($2n = 24$). Somatic chromosomes at metaphase. ($\times 3200$.)
3. *O. minuta* ($2n = 48$). Somatic chromosomes at metaphase. ($\times 3200$.)

[Continued at foot of next page.]

uncertain whether such products will be economically superior to the present rice species. At any rate it would be extremely interesting from both a scientific and an economic viewpoint to undertake further breeding work with this material.

A cytogenetical study of a large number of the species of *Oryza* obtained from different parts of the world has been undertaken, and already crosses have been made in the hope to create a synthetic species of *Oryza minuta* in the same way as Müntzing (1932) produced a synthetic *Galeopsis Tetrahit* and Kostoff (1936) created a synthetic wheat species, *Triticum Timococcum*, resistant to fungus diseases.

SUMMARY

1. The chromosomes of *O. officinalis* Wall. ($2n = 24$) have been examined and found to be larger in size than the

4. Semi-diagrammatic representation of the haploid genome of *O. minuta*, indicating the two sets of different sized chromosomes from the two 12-paired species.
5. *O. sativa*. Side view at metaphase I, with 12 smaller bivalents. ($\times 2300$.)
6. *O. officinalis*. Side view at metaphase I, with 12 larger bivalents. ($\times 2300$.)
7. Polar view at metaphase I, showing maximum secondary association, viz.: two groups of three bivalents and three groups of two bivalents of *O. officinalis*. ($\times 2300$.)
8. A giant pollen mother-cell at diakinesis with 24 bivalents in *O. officinalis*. ($\times 2300$.)
9. A 'Restitution Nucleus' with 24 univalents at metaphase I in *O. officinalis*. ($\times 2300$.)
10. Diakinesis in the hybrid *O. sativa* \times *O. officinalis*, with 12 small univalents and 12 larger univalents. ($\times 2300$.)
11. First anaphase in the hybrid *O. sativa* \times *O. officinalis*, showing irregular distribution of the univalents. ($\times 2300$.)
12. Metaphase I, in polar view showing maximum secondary association, viz.: 12 groups of two bivalents in each in *O. minuta*. ($\times 2300$.)
13. Polar view at metaphase II, showing varying degrees of secondary association of univalents in *O. minuta*. ($\times 2300$.)
14. Side view at metaphase I, in the hybrid *O. minuta* \times *O. officinalis*, showing 12 larger autosyndetic bivalents and 12 small univalents. ($\times 2300$.)
15. Side view at metaphase I, in the hybrid *O. minuta* \times *O. sativa*, showing 12 smaller autosyndetic bivalents and 12 larger univalents. ($\times 2300$.) N.B.—Chromosomes separated laterally.

chromosomes of *O. sativa* L. *O. officinalis* agrees with *O. sativa* in chromosome number, chiasma frequency, and degree and type of secondary association of the bivalents or univalents at metaphase I or II.

2. The phenomenon of pollen mother-cell fusion and formation of ' restitution nucleus ' were observed at meiosis in *O. officinalis*. These aberrations lead to diploid gametes formation with 24 chromosomes in each gamete.

3. The F_1 hybrid of *O. sativa* \times *O. officinalis* shows complete lack of synapsis and presence of univalents. The differences in chromosome size observed in the two parent species are maintained in the hybrid. Shrivelled pollen grains of different sizes and numbers are formed as a result of irregular meiotic divisions.

4. A detailed study of the somatic chromosomes of *O. minuta* Presl ($2n = 48$) shows that 24 chromosomes are small and 24 chromosomes are larger. These differences in the size of the chromosomes of *O. minuta* are comparable with the differences in size of the chromosomes of *O. sativa* and *O. officinalis*.

5. *O. minuta* is characterised by 24 bivalents at meiosis. Secondary pairing occurs among the markedly different sized bivalents or univalents at metaphase I or II. The maximum number of secondary association observed is 12 groups of two bivalents; 12 is therefore the basic number of *O. minuta*.

6. Of the interspecific hybrid *O. minuta* \times *O. officinalis* 12 bivalents are formed which are bigger in size. Pairing is autosyndetic, each of the bivalents being of similar size and shape. The rest of the 12 chromosomes are smaller in size than the bivalents and are non-homologous.

7. In the hybrid *O. minuta* \times *O. sativa* 12 bivalents are also formed which are smaller in size than the rest of the 12 univalents. The chromosome pairing is autosyndetic, each bivalent being of similar size and configuration.

8. A comparison shows that these differences in chromo-

some size in the two hybrids correspond to the differences in size of the two sets of chromosomes in *O. minuta*.

9. The chiasma frequency is the same in the pure species and the hybrids despite the differences in chromosome sizes.

10. The differences in the sizes of the chromosomes in the species of *Oryza* are due to genic and structural changes in the morphology of the chromosomes.

11. The conclusion is drawn that *O. minuta* is an allopolyploid (modified octoploid) originated from a cross between *O. officinalis* and *O. sativa* as a result of diploid gametes formation, but possessing morphologically dissimilar and non-homologous sets of chromosomes.

12. This theory of the hybrid origin of *O. minuta* is amply confirmed also by the following facts :

- (a) *O. minuta* is very closely related morphologically to *O. officinalis* and *O. sativa*.
- (b) It represents a recombination of characters from both of these species.
- (c) Geographical distribution of the two species *O. officinalis* and *O. sativa* gives ample opportunity for such hybridisation.
- (d) Diploid gametes are of frequent occurrence in both the species.
- (e) The somatic chromosome number of *O. minuta* Presl ($2n = 48$) has the sum of the somatic chromosome numbers of *O. officinalis* Wall. ($2n = 24$) and *O. sativa* L. ($2n = 24$).
- (f) The chromosome pairing is autosyndetic in meiosis, as a result of which *O. minuta* is fully fertile and breeds true to type.

I take this opportunity to express my heartfelt gratitude to Sir J. C. Bose, F.R.S., for his constant encouragement and for kindly providing me with full facilities for research works in his Institute. I must also accord my best thanks

to Prof. R. R. Gates, F.R.S., at whose genetical laboratory I was able to begin these investigations.

REFERENCES

- BABCOCK, E. B., and SWEZY, O.: 'The Chromosomes of *Crepis biennis* L. and *Crepis ciliata* C. Koch,' *Cytologia*, vol. 6 (1935), pp. 256-265.
- CLAUSEN, J.: 'Genetical and Cytological Investigations on *Viola tricolor* L. and *V. arvensis* Murr.,' *Hereditas*, vol. 8 (1926), pp. 1-157.
- COLLINS, J. L., and MANN, M. C.: 'Interspecific Hybrids in *Crepis* II' (a preliminary report on the results of hybridising *Crepis setosa* with *Crepis capillaris* and with *C. biennis*), *Genetics*, vol. 8 (1923), pp. 212-232.
- COLLINS, J. L., HOLLINGSHEAD, L., and AVERY, P.: 'Interspecific Hybrids in *Crepis* III, Constant fertile forms containing chromosomes derived from two species,' *Genetics*, vol. 14 (1929), pp. 305-320.
- DARLINGTON, C. D.: *Recent Advances in Cytology* (London, 1932).
- GATES, R. R.: 'The Stature and Chromosomes of *Oenothera gigas* de Vries,' *Arch. f. Zellforsch.*, vol. 3 (1909), pp. 525-552.
- KARPECHENKO, G. D.: 'Polyploid Hybrids of *Raphanus sativus* × *Brassica oleracea*,' *Bull. Appl. Bot.*, vol. 17 (1927), pp. 305-410.
- KIHARA, H., and ONO, T.: 'Chromosomenzahlen und systematische Gruppierung der *Rumex*-Arten,' *Zeitschr. f. wiss. Biologie*, Abt. B, Bd. 4 (1927), pp. 475-481.
- KOSTOFF, D.: 'Studies on Polyploid Plants. XI.—Amphidiploid *T. Timopheevi* Zhuk. × *T. monococcum* L.,' *C. R. Acad. Sci. U.S.S.R.*, vol. i (1936), pp. 37-41.
- LA COUR, L.: 'Improvement in Every-day Cytological Technique,' *Jour. Roy. Micros. Soc.*, vol. li (1931), pp. 119-126.
- MÜNTZING, A.: 'Cyto-genetic Investigations on Synthetic *Galeopsis Tetrahit*,' *Hereditas*, vol. xvi (1932), pp. 105-154.
- NANDI, H. K.: 'Cytological Investigations of Rice Varieties,' *Cytologia* (in the press). Thesis approved for the Degree of Doctor of Philosophy, University of London (1936 A).
- NANDI, H. K.: 'The Chromosome Morphology, Secondary Association and Origin of Cultivated Rice,' *Jour. of Genetics*, vol. 33 (1936 B), pp. 315-336.
- NANDI, H. K.: 'Effect of Temperature on the Formation of Diploid Pollen Grains in *O. sativa* L.,' *Abst. Procd. Ind. Sci. Congress*, 1937 (1936 c).
- NEWTON, W. C. F., and PELLEW, C.: 'Primula kewensis and its Derivatives,' *Jour. of Genetics*, vol. 20 (1929), pp. 405-467.
- ROSENBERG, O.: 'Die semiheterotypische Teilung und ihre Bedeutung für die Entstehung Verdoppelter chromosomenzahlen,' *Hereditas, Lund.*, vol. 8 (1927), pp. 305-38.
- RYBIN, V. A.: 'Über einen allotetraploiden Bastard von *Nicotiana Tabacum* × *Nicotiana sylvestris*,' *Ber. Deuts. Bot. Ges.*, vol. 37 (1929), pp. 385-394.
- SHARP, L. W.: *Introduction to Cytology*, Ed. 3 (1934), McGraw Hill Book Co., New York.
- SKOVSTED, A.: 'Cytological Studies in Cotton. II.—Two Interspecific Hybrids between Asiatic and the New World Cottons,' *Jour. of Genet.*, vol. 28 (1934), pp. 407-424.
- STÄHLIN, A.: 'Morphologische und Zytologische Untersuchungen an Gramineen,' *Wissensch. Archiv f. Landwirtsch., A. Pflanzenbau*, Bd. 1, (1929), pp. 330-398.
- VRIES, H. DE: 'Gruppenweise Artbildung,' vol. 8 (1913), p. 365.

- WETTSTEIN, F. v.: 'Die Erscheinung der Heteroploidie, besonders in Pflanzenreich,' *Ergebnisse der Biologie*, Bd. 2 (1927), pp. 311-356.
- WETTSTEIN, F. v.: 'Morphologie und Physiologie des Formwechsels der Moose auf genetischer Grundlage. II.—Bibliotheca,' *Genetica*, Bd. 10, (1928), pp. 1-216.
- WETTSTEIN, F. v.: 'Bastardpolyploidie als Artbildungsvorgang bei Pflanzen,' *Die Naturwissenschaften*, Bd. 20 (1932), pp. 981-984.
- WINGE, O.: 'The Chromosomes—their Numbers and General Importance,' *Compt. rend. Trav. Lab., Charlsherg*, vol. 13 (1917), pp. 131-275.
- WINGE, O.: 'A Case of Amphidiploidy within the Collective Species *Erophila verna*,' *Hereditas*, vol. 18 (1932), pp. 181-191.
- WINKLER, H.: 'Über die experimentalle Erzeugung von Pflanzen mit abweichenden Chromosomenzahlen,' *Zeitschr. f. Botanik*, vol. 8 (1916), pp. 417-531.

X.—THE RACIAL AFFINITIES OF THE INHABITANTS OF THE RAJMAHAL HILLS

BY

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THE racial affinities of the Orāons of Chota Nagpur were discussed by the late Dr. P. C. Basu in the TRANSACTIONS, Vol. IX, of the Bose Research Institute. The present paper, though not dealing with any people of the same geographical region, attempts to analyse the somatic traits of a tribe hitherto very little known, but said to have some common features with the Orāons, as well as possessing a Dravidian tongue. The dissimilarity in the cultures of the two tribes is, however, very marked. The Orāons have a long list of exogamous totemic clans, whereas the Mālér have no such clans. If they did possess a clan system as a result of acculturation with the Orāons, its disappearance is a matter which has yet to be proved.

The Mālér referred to above are the true hillmen of the Rajmahal Hills. They are also known as the Sāoriā Pāhāriās. The Mālpāhāriās dwelling in the south of the Mālér country belong to all intents and purposes to a separate tribe ; that the two tribes belong to the same ethnic stock will, however, be evident from the observations given below. The Mālér at present reside on the hill-tops occupying the north-eastern portion of the district of Santal Perganas, where they are mainly found in Rajmahal, Pakur and Godda subdivisions. The Mālpāhāriās, on the other hand, live mostly in the plains occupying the whole of Dumka subdivision and part of southern Pakur. The

changes that are noticeable in the latter are undoubtedly due to their contact with the Hindus, with whom they are coming into closer contact in the plains. The above two tribes are to a certain extent seen mixed up both on the hills and in the plains. It thus happens that a large number of the Mālér are found residing in the plains, though the number of the Mālpāhāriās residing on the hills is comparatively few. The Mālpāhāriās living on the hills present



FIG. 38. Front view.



FIG. 39. Side view.

Hill Mālér. Stature, 1566 mm. ; C.I., 73·94 ;
N.I., 89·80 ; eye colour, 3.

some important features, suggesting that they occupy a stage of transition which has converted the Hill Mālér into the Mālpāhāriās. The true types of the Hill Mālpāhāriās¹ are at present met with only in the villages under Parerkola Bungalow in Pakur subdivision. In what follows, by 'Rajmahal aborigines' will be indicated the Hill Mālér, the Plains Mālér and the Mālpāhāriās.

The people whom I measured were taken from nearly all the centres of the Mālér and Mālpāhāriā countries. They have been grouped under the following three heads :

¹ The number of the Hill Mālpāhāriās being only 6, all the Mālpāhāriās are grouped under one head.

1. Measurements of the Mālér residents on the hills, to be distinguished as Hill Mālér residents (figs. 38, 39).



FIG. 40. Front view.



FIG. 41. Side view.

Plains Mālér. Stature, 1630 mm.; C.I., 82.22;
N.I., 86.67; eye colour, 2.

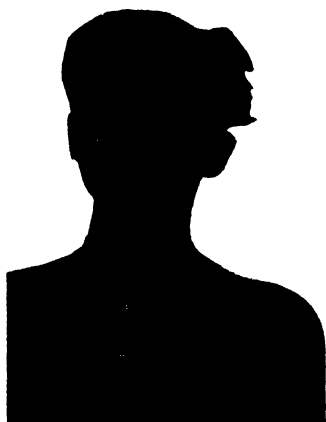


FIG. 42. Front view.



FIG. 43. Side view.

Mālpāhārīā. Stature, 1635 mm.; C.I., 73.80;
N.I., 102.50; eye colour, 3.

2. Measurements of the Mālér residents on the plains, specified as Plains Mālér residents (figs. 40, 41).
3. Measurements of the Mālpāhārīās (figs. 42, 43).

All these three types have been compared with the Orāons described by the late Dr. Basu.¹ The measurements are taken according to the technique advocated by the International Agreement of 1909 and its subsequent modifications. The subjects measured were all above 21 years of age, measurements of senile and deformed persons being avoided.

Analysis of the Anthropometric Data

Altogether 296 male individuals were measured, of which the number of the Hill Mālér's was 188, and that of the Plains Mālér's and the Mālpāhārīās 54 each.

A. PHYSICAL OBSERVATIONS

Integumentary Colours

The skin colour was examined against Von Luschan's Hautfarbentafel, on the forehead, on the ventral side of the upper arm and the chest. Table I gives a summary of the observations.

From that table it will be seen that in all the three groups of the Rajmahal aborigines No. 30 is the prevailing colour of the forehead. It is found in 52·41 per cent. among the Hill Mālér's, 58·49 per cent. among the Plains Mālér's and 43·48 per cent. among the Mālpāhārīās. The mean colour of the ventral side of the upper arm and chest is No. 32 in all cases. It varies between 31·91 per cent. and 30·81 per cent. among the Hill Mālér's for the upper arm and chest respectively; 37·74 per cent. for both the upper arm and chest among the Plains Mālér's; and 58·70 per cent. and 60·87 per cent. among the Mālpāhārīās for the upper arm and chest respectively. This shows that all the three groups possess the same skin colour. The colour of the forehead of the Orāons, according to Basu, is No. 27,

¹ P. C. Basu, 'The Racial Affinities of the Oraons,' *Trans. Bose Res. Inst.*, vol. ix, pp. 132-176.

TABLE I.—SKIN COLOUR
(After Von Luschan)

which is present in 64·80 per cent. ; the colours of the upper arm and the chest are Nos. 29 and 27 respectively, and the percentages are 36·80 and 54·00 respectively. This shows that while the skin colour of both are dark chocolate brown, that of the Orāons possesses a slightly lighter shade.

TABLE II.—HAIR
(After Eugen Fischer)

Scale	Hill Mālér's		Plains Mālér's		Mālpāhārīās	
	Freq.	Perct.	Freq.	Perct.	Freq.	Perct.
<i>Colour</i>						
27 . .	171	90·96	50	92·59	35	76·09
27 . . (Reddish)	—	—	1	1·85	—	—
28 . .	15	7·98	—	—	11	23·91
29 . .	2	1·06	3	5·56	—	—
<i>Character</i>						
Wavy . .	176	93·62	49	90·74	37	80·43
Curly . .	8	4·26	2	3·70	1	2·17
Straight . .	4	2·13	3	5·56	8	17·39
<i>Quantity</i>						
Moderate . .	105	55·85	41	75·93	38	82·69
Rich . .	83	44·15	13	24·75	8	17·39

TABLE III.—EYE

Scale	Hill Mālér's		Plains Mālér's		Mālpāhārīās	
	Freq.	Perct.	Freq.	Perct.	Freq.	Perct.
<i>Colour (after Martin)</i>						
I . .	12	6·38	I	1·85	I	2·17
2 . .	60	31·91	16	29·63	7	15·22
3 . .	90	47·87	25	46·30	25	54·35
3 (Bluish)	—	—	I	1·85	—	—
4 . .	25	13·30	11	20·37	13	28·26
6 ¹ . .	I	0·53	—	—	—	—
<i>Form of the Eye-slit</i>						
Straight .	159	84·57	51	94·44	41	89·13
Oblique .	29	15·43	3	5·56	5	10·87
<i>Epicanthic Fold</i>						
Nil . .	149	79·26	43	79·63	38	82·69
Traces in :—						
Right Eye .	8	4·26	—	—	—	—
Left Eye .	3	1·60	—	—	—	—
Both Eyes .	28	14·89	11	20·37	8	17·39

¹ See figs. 44 and 45.

The hair was examined with the help of the Haarfarbentafel of Fischer. In all the three groups the colour is black corresponding to No. 27 of the scale. The form of the hair is wavy, and Table II shows the distribution of the quantity of the hair. In both the form and the colour of the hair the Rajmahal Hillmen agree with the Orāons.

The average colour of the iris is dark brown, as was found by Martin's Augenfarbentafel, and corresponds to

FIG. 44. Front view.

FIG. 45. Side view.

Hill Mālér. Stature, 1550 mm. ; C.I., 73·22 ;
N.I., 93·62 ; eye colour, 6.

No. 3 of the above scale. This was found in the following percentages among the three groups : Hill Mālér 47·87, Plains Mālér 46·30, and Mālpāhāriās 54·35. The colour of the iris of the Orāons is dark brown, the majority possessing eyes corresponding to No. 2 of the scale and its percentage is 80·80. The eye colour corresponding to No. 2, as seen in Table III, comes second in both the percentages and the frequencies among the Mālér ; but it should, however, be noted that No. 4 is also fairly distributed among the Rajmahal aborigines, whereas the latter is insignificant among the Orāons. It is interesting to note that darker eye

colours are found frequently to be associated with denser jungles and higher altitudes.

The form of the eye-slit in all the three groups is horizontal, but, as will have been noticed in Table III, there is a small percentage of people with oblique eyes—the percentage being the greatest among the Mālér's (15·43). Among the Orāons the percentage of people with horizontal eyes as noticed by Basu is 65·20, and that of the oblique eyes is 34·80. It will be noted, therefore, that in comparison with the Mālér's, the Orāons show a very much larger percentage of people with oblique eyes.

TABLE IV.—FOREHEAD

Character	Hill Mālér's		Plains Mālér's		Mālpāhārīās	
	Freq.	Perct.	Freq.	Perct.	Freq.	Perct.
<i>Height</i>						
Moderate .	64	34·04	16	29·63	10	21·74
Pronounced .	124	65·96	38	70·37	36	78·27
<i>Breadth</i>						
Moderate .	63	33·51	18	33·33	11	23·91
Pronounced .	125	66·49	36	66·66	35	76·09
<i>Slope Backwards</i>						
Nil . .	98	52·13	12	22·22	10	21·74
Moderate .	64	34·04	32	59·26	15	32·69
Pronounced .	26	13·83	10	18·52	21	45·65

Both the height and the breadth of the forehead are pronounced among all the three groups of the Rajmahal aborigines. The backward slope of the forehead is more marked among the Mālpāhāriās than the two Mālér groups, but among the Plains Mālér it is more developed than the Hill Mālér.

TABLE V.—SUPRAORBITAL RIDGES

Character	Hill Mālér		Plains Mālér		Mālpāhāriās	
	Freq.	Perct.	Freq.	Perct.	Freq.	Perct.
Nil . .	23	19·15	—	—	2	4·35
Moderate .	129	68·62	37	68·52	37	80·43
Pronounced .	36	12·23	17	31·48	7	15·22

The supraorbital ridges of all the three groups of Rajmahal aborigines are moderate, though there is an appreciable percentage of people having strongly developed ridges.

TABLE VI.—ALVEOLAR PROGNATHISM

Character	Hill Mālér		Plains Mālér		Mālpāhāriās	
	Freq.	Perct.	Freq.	Perct.	Freq.	Perct.
Nil . .	142	77·60	31	57·41	39	84·78
Moderate .	34	18·58	17	31·48	4	8·70
Pronounced .	7	3·93	6	11·11	3	6·52

The majority of the people examined do not show any alveolar prognathism, but among the two Mālér groups there is a section which exhibits moderate development. This is, however, more marked among the two Mālér groups than among the Mālpāhāriās.

TABLE VII.—NOSE

Character	Hill Mālér's		Plains Mālér's		Mālpāhārīās	
	Freq.	Perct.	Freq.	Perct.	Freq.	Perct.
<i>Nasion Depression</i>						
Shallow . .	133	70·74	31	57·41	31	67·39
Deep . .	55	29·26	23	42·60	15	32·61
<i>Nasal Profile</i>						
Straight .	46	24·47	12	22·22	11	23·91
Concave .	68	36·17	20	37·04	26	56·52
Convex . .	11	5·85	3	5·56	1	2·17
Flat . .	63	33·51	19	35·19	8	17·39
<i>Nasal Tip</i>						
Inclination :—						
Upwards .	57	30·32	19	35·19	22	47·83
Downwards	45	23·94	13	24·75	8	17·39
Horizontal .	86	45·74	22	40·74	16	34·78

The nasion is slightly depressed in all the three groups. A marked depression, however, was observed among a greater percentage of people with flat broad noses.

The bridge of the nose is in general concave in all the three groups, but stray cases of what appeared to be flat nose occurred in all the three sections.

The nasal septum is usually horizontal among the two

Mālér groups, but among the Mālpāhāriās it is not so. The inclination of the tip of the nose is upwards among the majority of the three groups; there is, however, an appreciable number in all the groups where it is tilted downwards.

FIG. 46. Front view.

FIG. 47. Side view.

Hill Mālér. Stature, 1566 mm.; C.I., 71·98;

N.I., 90·70; eye colour, 3.

TABLE VIII.—LIPS

Character	Hill Málér		Plains Málér		Mālpāhāriās	
	Freq.	Perct.	Freq.	Perct.	Freq.	Perct.
Thin . .	1	0·53	1	1·85	1	2·17
Medium .	128	68·09	25	46·30	36	78·27
Thick . .	10	5·31	—	—	1	2·17
Everted .	49	26·06	28	51·85	8	17·39

The lip is medium and not thick among all the three groups; there is a larger percentage of people among the

Mālér's, especially the Plains Mālér's, who have everted lips (51·85 per cent.). Among the Mālpāhārīās the frequency is much less (17·39 per cent.).

TABLE IX.—CHIN

Character	Hill Mālér's		Plains Mālér's		Mālpāhārīās	
	Freq.	Perct.	Freq.	Perct.	Freq.	Perct.
Round . .	79	42·47	25	46·30	18	39·13
Pointed . .	60	32·26	15	27·78	13	28·26
Square . .	39	20·97	10	18·52	14	30·43
Bifid square .	1	0·54	1	1·85	1	2·17
Bifid round .	7	3·76	2	3·70	—	—

The chin is round in the majority of cases ; a considerable number, however, possess chins which are pointed.

TABLE X.—FACE

Character	Hill Mālér's		Plains Mālér's		Mālpāhārīās	
	Freq.	Perct.	Freq.	Perct.	Freq.	Perct.
Square . .	54	28·72	17	31·48	19	41·30
Pear . .	9	4·79	2	3·70	—	—
Oval . .	36	19·15	5	9·26	4	8·70
Round . .	7	3·72	5	9·26	2	4·35
Triangular .	34	18·09	10	18·52	8	17·39
Long . .	48	25·53	15	27·78	13	28·26



FIG. 48. Front view.



FIG. 49. Side view.

Hill Málé. Stature, 1483 mm.; C.I., 71·95;
N.I., 82·00; F.I., 90·70; eye colour, 3.



FIG. 50. Front view.



FIG. 51. Side view.

Málpāhāriā. Stature, 1637 mm.; C.I., 74·74;
N.I., 62·96; F.I., 93·18; eye colour, 3.

The face is, generally speaking, squarish in shape among all the three groups; there is, however, an appreciable percentage of people with a triangular type of face.

The disharmonic type of face is common—the percentages being 14·43, 20·37 and 22·22 among the Hill Mālér's (figs. 48, 49), the Plains Mālér's and the Mālpāhārīās (figs. 50, 51) respectively.

B. ANTHROPOMETRIC MEASUREMENTS

Stature

The average stature of the Rajmahal aborigines is short, the mean statures of the Hill Mālér's, the Plains Mālér's and the Mālpāhārīās being $1566\cdot40 \pm 2\cdot55$ mm., $1581\cdot80 \pm 4\cdot60$ mm. and $1573\cdot60 \pm 5\cdot31$ mm. respectively. The Orāons, on the other hand, are somewhat taller, and according to Basu the mean stature is $1618\cdot00 \pm 2\cdot23$ mm., which is 51·60 mm., 36·20 mm. and 44·40 mm. higher than the mean statures of the Hill Mālér's, the Plains Mālér's and the Mālpāhārīās respectively. The higher stature of the Plains Mālér's seems to be due to better means of living. An instance of it can be seen in the Mettur Pahar village under Kusma Bungalow in Rajmahal subdivision. This village is inhabited by the Hill Mālér's and a small group has settled down on a lower altitude from the main hill. Individuals of this latter place are found to be taller in stature than the peoples of the main hill. It should be mentioned here that the Plains Mālér's have at present better means of livelihood than those of the other two groups. They have begun to own lands at the foot of the hills, and most of them have taken to cultivation with the plough. In the above village flourishing vegetable gardens are to be found, being the first of their kind among these people. The following table shows the difference in the statures of the above three groups :

TABLE XI.—STATURE
(After Martin)

Character	Hill Mālér's		Plains Mālér's		Mālpāhāriās	
	Freq.	Perct.	Freq.	Perct.	Freq.	Perct.
Short . .	152	80.42	36	66.67	36	66.67
Medium .	37	19.58	17	31.48	17	31.48
Tall . .	—	—	1	1.85	1	1.85

The general results are illustrated in the diagrammatic curves given in fig. 52. The greatest frequency of the stature of the Rajmahal Hill tribes falls between 1554 mm. and 1614 mm., showing that the majority are of short stature.

Length Breadth Index

Like most of the aboriginal tribes of Central India the Rajmahal groups are mainly dolichocephalic.

TABLE XII

Character	Hill Mālér's		Plains Mālér's		Mālpāhāriās	
	Freq.	Perct.	Freq.	Perct.	Freq.	Perct.
Dolichocephal	134	70.90	37	68.52	37	68.52
Mesocephal .	53	28.04	15	27.78	16	29.63
Brachycephal	2	1.06	2	3.70	1	1.85

The mean cephalic indices of the Hill Mālér's, the Plains Mālér's and the Mālpāhāriās are 74.54 ± 0.13 , 74.65 ± 0.29 and 74.54 ± 0.26 , the standard deviations being 2.69 ± 0.09 , 3.12 ± 0.20 and 2.82 ± 0.18 respec-

tively. The mean cephalic lengths are 184.21 ± 0.27 , 184.54 ± 0.50 and 184.30 ± 0.58 respectively, the

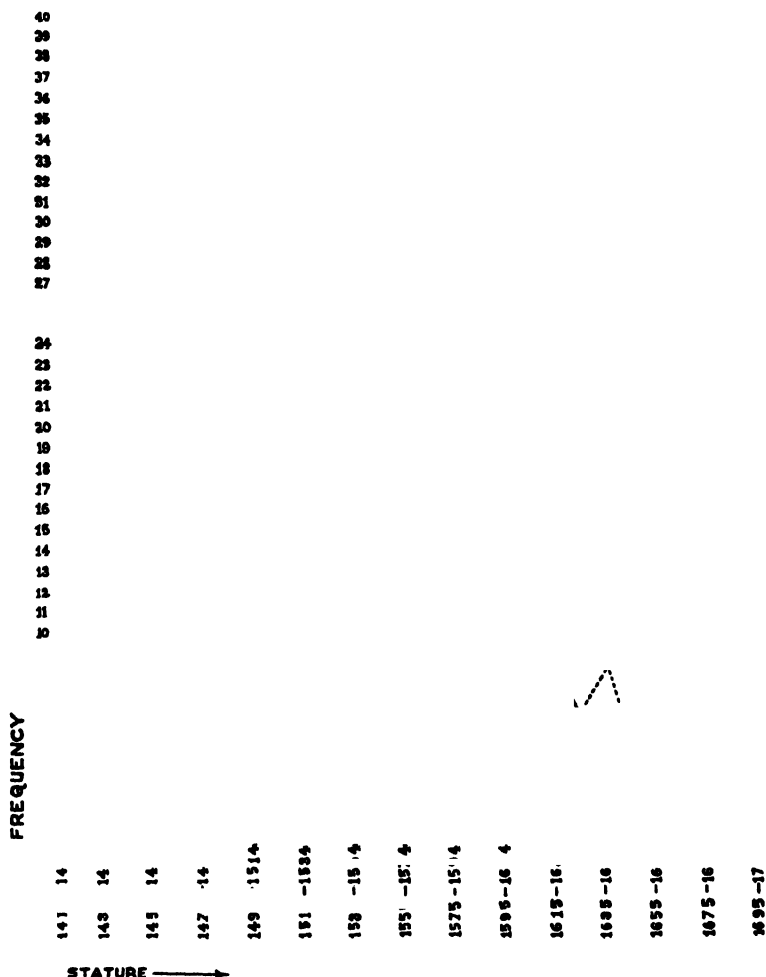


FIG. 52. Frequency curve of stature.
 — Hill Mälé. — Plains Mälé. - - - - - Mälpähäriä.

standard deviations being 5.61 ± 0.20 , 5.41 ± 0.35 and 6.27 ± 0.41 . The mean cephalic breadths are 137.23 ± 0.20 , 137.70 ± 0.38 and 137.11 ± 0.39 , with standard

deviations of 4.16 ± 0.14 , 4.13 ± 0.27 , and 4.28 ± 0.28 respectively. In regard to the Orāons the mean cephalic index, according to Basu, is 74.32 ± 0.13 , with the standard deviation of 3.06 ± 0.09 ; the mean cephalic breadth is 138.20 ± 0.17 , the standard deviation being 3.99 ± 0.17 .

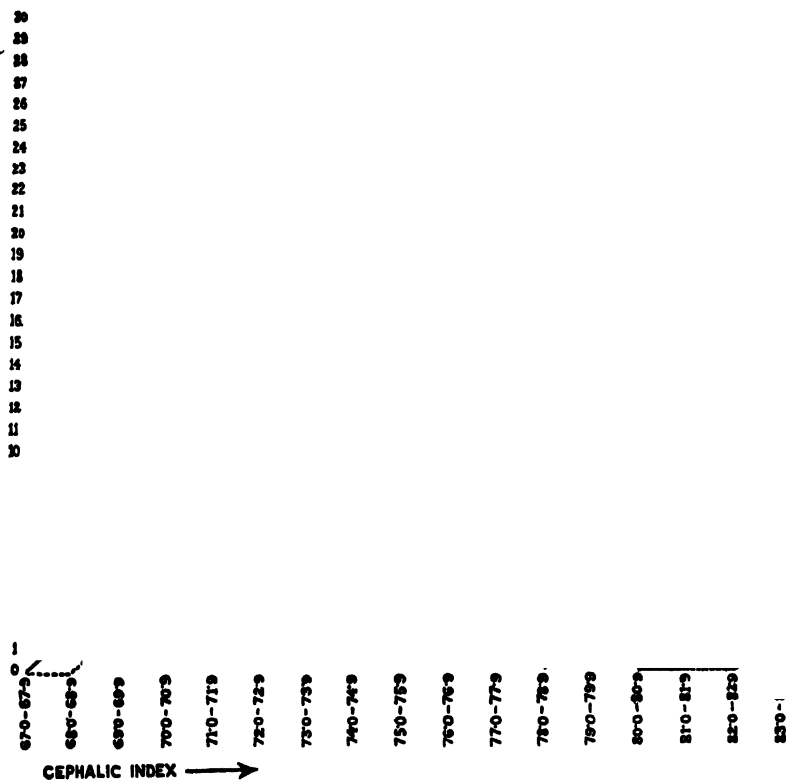


FIG. 53. Frequency curve of cephalic index.
 — Hill Mālér. — Plains Mālér. - - - - Mālpāhārīā.

The Orāons therefore possess a slightly longer head. As will be noticed from fig. 53, the greatest frequency of the cephalic index of the Mālér falls between 73 and 77 and that of the Mālpāhārīās between 73 and 76, and that of the Plains Mālér is between 72 and 76. All these show the definitely elongated nature of the head.

TABLE XIII.—LENGTH HEIGHT INDEX

Character	Hill Mālér's		Plains Mālér's		Mālpāhārīās	
	Freq.	Perct.	Freq.	Perct.	Freq.	Perct.
Chamaecephal	—	—	—	—	—	—
Orthocephal .	—	—	—	—	—	—
Hypsicephal .	187	100	54	100	54	100

All the three groups of the Rajmahal aborigines are predominantly and uniformly hypsicephalic. The mean indices of the Hill Mālér's, the Plains Mālér's and the Mālpāhārīās are 72.17 ± 0.18 , 71.26 ± 0.33 and 70.61 ± 0.36 respectively, the respective standard deviations being 3.36 ± 0.11 , 3.58 ± 0.23 and 3.88 ± 0.25 .

TABLE XIV.—BREADTH HEIGHT INDEX

Character	Hill Mālér's		Plains Mālér's		Mālpāhārīās	
	Freq.	Perct.	Freq.	Perct.	Freq.	Perct.
Tapeinocephal	—	—	—	—	—	—
Metriocephal .	2	1.06	1	1.85	—	—
Acrocephal .	186	98.99	53	98.15	54	100

As in the length height index, the Rajmahal aborigines are also predominantly acrocephalic in character. The mean indices of the Hill Mālér's, the Plains Mālér's and the Mālpāhārīās are 96.82 ± 0.24 , 95.19 ± 0.48 and 95.02 ± 0.45 , with the respective standard deviations as 4.88 ± 0.17 , 5.23 ± 0.34 and 4.87 ± 0.32 .

TABLE XV.—ORBITONASAL INDEX

Character	Hill Mālér's		Plains Mālér's		Mālpāhārīās	
	Freq.	Perct.	Freq.	Perct.	Freq.	Perct.
Platyopic .	71	39·44	26	48·15	20	43·48
Mesopic .	53	29·44	16	29·63	11	23·91
Pro-opic .	56	31·11	12	22·22	15	32·69

The mean orbitonasal indices of the Hill Mālér's, the Plains Mālér's and the Mālpāhārīās are $111·31 \pm 0·23$, $110·17 \pm 0·32$ and $110·92 \pm 0·36$, with the respective standard deviations as $4·55 \pm 0·17$, $3·51 \pm 0·23$ and $3·61 \pm 0·26$, which show that all of them are platyopic. Compared with the figures of the Orāons as published by Basu, it would appear that the Rajmahal aborigines have a flatter face.

TABLE XVI.—NASAL INDEX

Character	Hill Mālér's		Plains Mālér's		Mālpāhārīās	
	Freq.	Perct.	Freq.	Perct.	Freq.	Perct.
Leptorrhine .	5	2·65	—	—	5	9·26
Mesorrhine .	100	52·91	28	51·85	27	50·00
Platyrrhine .	74	39·15	25	46·30	20	37·37
Hyper-platyrrhine	10	5·29	1	1·85	2	3·70

The mean nasal indices of the Hill Mālér's, the Plains Mālér's and the Mālpāhārīās are $84·30 \pm 0·43$, $84·62 \pm 0·63$ and $79·10 \pm 0·76$ respectively, with the standard deviations of $8·70 \pm 0·30$, $6·88 \pm 0·45$ and $8·32 \pm 0·54$ respectively. As will be noticed from the above table and

the frequency curves given in fig. 54, the two Mālér groups are virtually platyrrhine. The Mālpāhāriās, however, have a much lower index. When we compare the percentage distribution, it will be seen that the largest groups are mesorrhine in all the three tribes, the frequencies for the platyrrhine group being only slightly less. It may therefore be concluded that all the Rajmahal tribes have broad noses, almost platyrrhine. The Orāon nose, according to Basu, is also 'inclined to be broad,' its mean nasal index being 82.71 ± 0.28 .

Nasal Elevation Index

The mean nasal elevation indices of the Hill Mālér, the Plains Mālér and the Mālpāhāriās are 49.28 ± 0.17 , 50.04 ± 0.51 and 51.57 ± 0.49 respectively, with the standard deviations of 5.44 ± 0.19 , 5.53 ± 0.36 and 4.96 ± 0.35 respectively. The author has attempted a classification of the above index on the basis of a series of data obtained by Dr. B. S. Guha during the last Census operations, and has proposed the following classification :

Platysept	.	.	.	49.9
Mesosept	.	.	.	50.0 — 64.9
Hypsisept	.	.	.	65.0 +

Applying the above classification, the Rajmahal aborigines may be classed as follows :

TABLE XVII.—NASAL ELEVATION INDEX

Character	Hill Mālér		Plains Mālér		Mālpāhāriās	
	Freq.	Perct.	Freq.	Perct.	Freq.	Perct.
Platysept .	95	50.80	27	50.00	15	32.61
Mesosept .	92	49.20	27	50.00	31	67.39
Hypsisept .	—	—	—	—	—	—

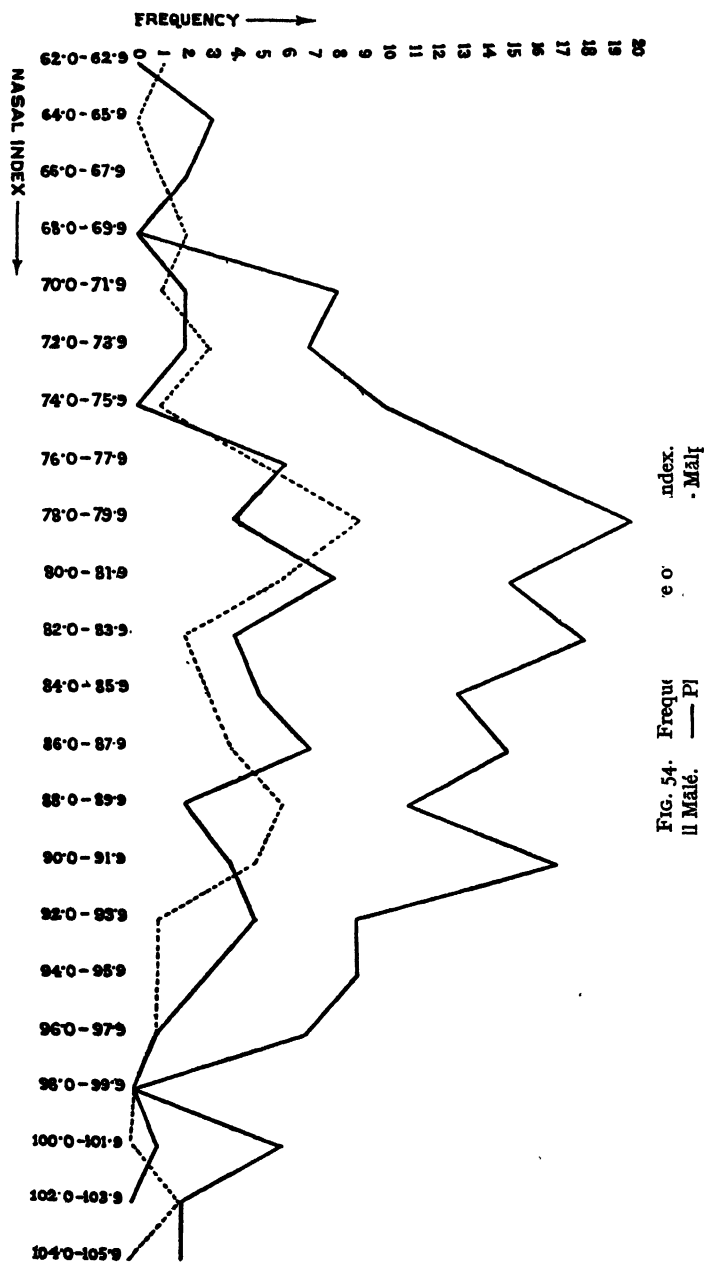


FIG. 54. Frequencies of Nasal Index in Malé.

All the three groups therefore possess the highest percentages of platysept noses; the high percentages of the mesosept character are also quite in accord with the high percentages of the mesorrhine noses (Table XVI) in all the three groups. As will be seen from the frequency curves given in fig. 55 this evenness of distribution is marked in all the three groups and the very close association of the three groups in this particular character.

Upper Facial Index

Of the Rajmahal aborigines the Mālér's have the highest percentage of medium faces, while the Mālpāhārīās possess an equal percentage of medium and long faces.

TABLE XVIII.—UPPER FACIAL INDEX

Character	Hill Mālér's		Plains Mālér's		Mālpāhārīās	
	Freq.	Perct.	Freq.	Perct.	Freq.	Perct.
Hyper-euryene	3	1.59	1	1.85	—	—
Euryene .	53	28.04	21	38.89	2	4.35
Mesene . .	89	47.09	23	42.59	17	36.96
Leptene .	30	15.87	8	14.82	17	36.96
Hyper-leptene	14	7.41	1	1.85	10	21.74

The mean upper facial indices of the Hill Mālér's, the Plains Mālér's and the Mālpāhārīās are 50.18 ± 0.19 , 49.12 ± 0.34 and 54.11 ± 0.38 respectively, with the standard deviations as 3.95 ± 0.14 , 3.67 ± 0.24 and 3.83 ± 0.27 .

Total Facial Index

The Hill Mālér's have the highest percentage of flat (Euryprosopic) faces, while both the Plains Mālér's and the

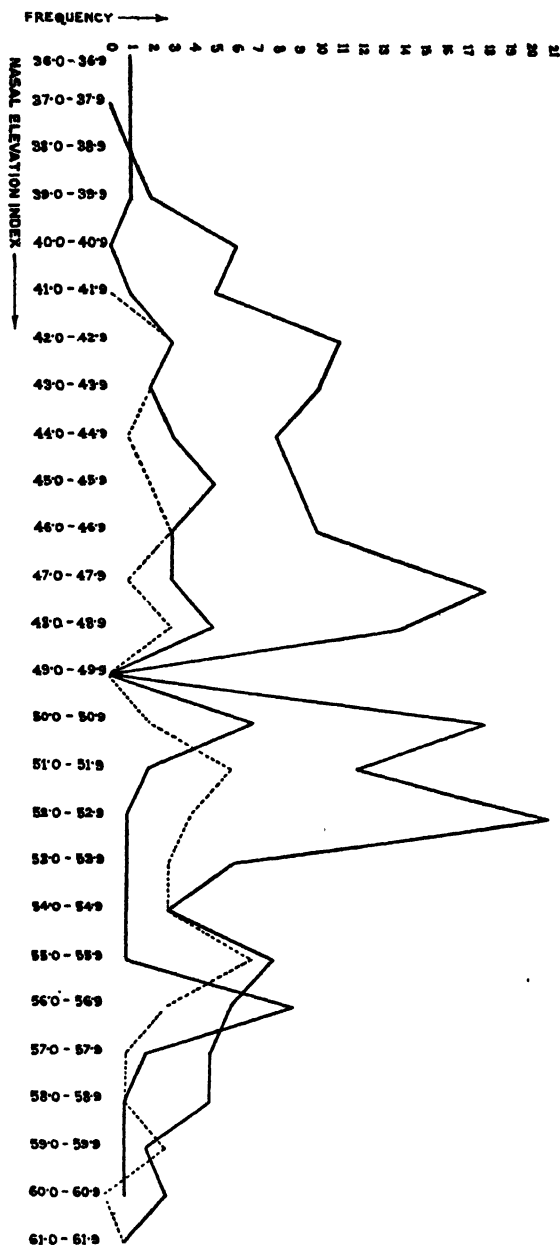


FIG. 55. Frequency curve of nasal elevation index.
 — Hill Malé. — Plains Malé. - - - - Malpáharia.

Mālpāhāriās possess the largest percentage of medium (mesoprosopic) faces. Basu found the Orāons to be long (leptoprosopic).

TABLE XIX.—TOTAL FACIAL INDEX

Character	Hill Mālér's		Plains Mālér's		Mālpāhāriās	
	Freq.	Perct.	Freq.	Perct.	Freq.	Perct.
Hyper-euryprosopic	30	15·87	11	20·37	9	16·67
Euryprosopic .	67	35·45	12	22·22	13	24·75
Mesoprosopic .	59	31·22	16	29·63	18	33·33
Leptoprosopic	30	15·87	12	22·22	11	20·37
Hyper-leptoprosopic	3	1·59	3	5·56	3	5·55

Taking the face as a whole, the difference in the three groups appears to be insignificant, as the euryprosopic character is dominant in all. The mean total facial indices of the Hill Mālér's, the Plains Mālér's and the Mālpāhāriās are $83·74 \pm 0·22$, $84·61 \pm 0·48$ and $84·94 \pm 0·47$ respectively, the standard deviations being respectively $4·65 \pm 0·16$, $5·27 \pm 0·34$ and $5·07 \pm 0·33$.

The statistical constants of the measurements and indices of the Rajmahal aborigines are given below in Table XX.

CONCLUSION

The results obtained show that the only difference that can be seen in the three groups of Rajmahal aborigines lies in the stature, which is most probably due to their change of habitats. The difference between the Hill Mālér's and

TABLE XX.—STATISTICAL CONSTANTS OF THE

Measurements and Indices	Hill Malers			
	No.	Mean with P.E.	S.D. with P.E.	Coefficient of Variation with P.E.
Stature	189	1566.40 \pm 2.55	52.00 \pm 1.80	3.32 \pm 0.13
Auricular height	188	133.52 \pm 0.33	6.72 \pm 0.23	5.03 \pm 0.18
Maximum head length	189	184.21 \pm 0.27	5.61 \pm 0.20	3.05 \pm 0.11
Maximum head breadth	189	137.23 \pm 0.20	4.16 \pm 0.14	3.03 \pm 0.11
Minimum frontal breadth	187	100.54 \pm 0.17	3.47 \pm 0.12	3.45 \pm 0.12
Maximum bizygomatic breadth	189	130.98 \pm 0.22	4.41 \pm 0.15	3.37 \pm 0.12
Bigonial breadth	188	96.89 \pm 0.28	5.67 \pm 0.20	5.85 \pm 0.20
Interorbital breadth	187	33.57 \pm 0.16	3.29 \pm 0.11	9.80 \pm 0.35
Orbitonasal breadth	188	101.53 \pm 0.21	4.37 \pm 0.15	4.30 \pm 0.15
Orbitonasal arc	182	112.65 \pm 0.26	5.17 \pm 0.18	4.59 \pm 0.16
Nasal length	188	47.20 \pm 0.17	3.43 \pm 0.12	7.27 \pm 0.25
Nasal breadth	189	39.61 \pm 0.14	2.87 \pm 0.10	7.25 \pm 0.25
Nasal height	188	19.49 \pm 0.10	2.12 \pm 0.08	10.88 \pm 0.38
Upper facial length	189	65.68 \pm 0.26	5.34 \pm 0.19	8.13 \pm 0.28
Total facial length	189	109.67 \pm 0.28	5.71 \pm 0.20	5.21 \pm 0.19
Horizontal circumf. of the head	91	532.05 \pm 1.28	13.40 \pm 0.67	2.52 \pm 0.13
Sagittal arc	93	328.95 \pm 1.32	13.70 \pm 0.69	4.16 \pm 0.21
Transverse arc	94	328.40 \pm 1.37	14.10 \pm 0.69	4.29 \pm 0.21
Length breadth index	189	74.54 \pm 0.13	2.69 \pm 0.09	3.61 \pm 0.13
Length height index	187	72.17 \pm 0.18	3.36 \pm 0.11	4.66 \pm 0.16
Breadth height index	188	96.82 \pm 0.24	4.88 \pm 0.17	5.04 \pm 0.18
Orbitonasal index	180	111.31 \pm 0.23	4.55 \pm 0.17	4.09 \pm 0.15
Nasal index	189	84.30 \pm 0.43	8.70 \pm 0.30	10.32 \pm 0.36
Nasal elevation index	187	49.28 \pm 0.17	5.44 \pm 0.19	11.04 \pm 0.39
Upper facial index	189	50.18 \pm 0.19	3.95 \pm 0.14	7.87 \pm 0.28
Total facial index	189	83.74 \pm 0.22	4.65 \pm 0.16	5.55 \pm 0.19
Trans. cephalofacial index	188	97.84 \pm 0.15	3.01 \pm 0.10	3.08 \pm 0.11
Vertical cephalofacial index	188	82.48 \pm 0.25	5.10 \pm 0.18	6.18 \pm 0.22

MEASUREMENTS AND INDICES OF RAJMAHAL ABORIGINES

Plains Málérá				Málpáhará			
No.	Mean with P.E.	S.D. with P.E.	Coefficient of Variation with P.E.	No.	Mean with P.E.	S.D. with P.E.	Coefficient of Variation with P.E.
54	1581.80±4.60	50.20±3.25	3.17±0.21	54	1573.60±5.31	57.80±3.75	3.67±0.24
54	131.30±0.58	6.34±0.41	4.83±0.31	54	129.96±0.60	6.57±0.43	5.06±0.16
54	184.54±0.50	5.41±0.35	2.93±0.19	54	184.30±0.58	6.27±0.41	3.40±0.07
54	137.70±0.38	4.13±0.27	3.00±0.19	54	137.11±0.39	4.28±0.28	3.12±0.06
53	100.15±0.31	3.36±0.22	3.35±0.22	46	100.13±0.32	3.18±0.22	3.18±0.07
54	130.28±0.41	4.45±0.29	3.42±0.22	54	132.07±0.40	4.32±0.28	3.27±0.08
53	94.49±0.50	5.36±0.35	5.67±0.37	46	99.52±0.57	5.71±0.40	5.74±0.13
54	31.07±0.20	2.13±0.14	6.86±0.45	46	31.63±0.25	2.51±0.18	7.94±0.18
54	101.53±0.37	4.03±0.26	3.97±0.26	46	106.48±0.61	6.10±0.40	5.73±0.13
53	111.53±0.37	3.97±0.26	3.56±0.23	46	112.50±0.47	4.76±0.33	4.23±0.09
54	45.98±0.31	3.35±0.22	7.29±0.47	54	47.52±0.33	3.62±0.23	7.62±0.16
54	38.70±0.22	2.38±0.15	6.15±0.40	54	39.07±0.25	2.68±0.17	6.86±0.14
54	19.24±0.19	2.10±0.14	10.09±0.66	46	20.22±0.18	1.85±0.13	9.15±0.21
54	63.87±0.41	4.42±0.29	6.92±0.45	46	71.02±0.48	4.83±0.34	6.80±0.15
54	110.06±0.55	6.03±0.39	5.48±0.36	54	112.20±0.61	6.64±0.43	5.92±0.12
41	531.25±1.12	10.65±0.79	2.00±0.15	33	527.45±1.87	15.90±1.32	3.01±0.08
41	328.60±1.28	12.10±0.90	3.68±0.27	33	326.65±2.44	20.75±1.72	6.35±0.18
41	324.24±0.68	6.46±0.48	1.99±0.15	33	333.96±1.33	11.31±0.94	3.39±0.09
54	74.65±0.29	3.12±0.20	4.18±0.27	54	74.54±0.26	2.82±0.18	3.78±0.08
54	71.26±0.33	3.58±0.23	5.02±0.33	54	70.61±0.36	3.88±0.25	5.49±0.11
54	95.19±0.48	5.23±0.34	5.49±0.36	54	95.02±0.45	4.87±0.32	5.13±0.11
54	110.17±0.32	3.51±0.23	3.19±0.21	46	110.92±0.36	3.61±0.26	3.25±0.07
54	84.62±0.63	6.88±0.45	8.13±0.53	54	79.10±0.76	8.32±0.54	10.52±0.22
54	50.04±0.51	5.53±0.36	11.11±0.73	46	51.57±0.49	4.96±0.35	9.62±0.22
54	49.12±0.34	3.67±0.24	7.47±0.49	46	54.11±0.38	3.82±0.27	7.08±0.16
54	84.61±0.48	5.27±0.34	6.23±0.41	54	84.94±0.47	5.07±0.33	5.97±0.12
54	94.46±0.29	3.13±0.11	3.31±0.11	54	95.76±0.86	9.32±0.61	9.73±0.20
54	84.00±0.55	6.02±0.39	7.17±0.47	54	86.66±0.63	6.90±0.45	7.96±0.16

the Plains Mālér is considerable in the transverse arc and transverse cephalofacial index of the head ; slight differences occur, however, in auricular height, in bigonial breadth, and in interorbital breadth. The difference between the Hill Mālér and the Mālpāhāriās is great in auricular height, in orbitonasal breadth, in upper facial length, in horizontal circumference of head, in transverse arc of head, in nasal index, and in vertical cephalofacial index. There are, however, slight differences in bigonial breadth, in total facial length, in sagittal arc of the head, nasal elevation index, and in transverse cephalofacial index. The above shows that the Mālpāhāriās differ more from the Hill Mālér than the latter do from the Plains Mālér. It would appear that the Mālpāhāriās must have separated a very long time ago and that the changes, due to the environments of the plains, have considerably influenced them.

From the above results it will be seen that the three groups of Rajmahal aborigines are so closely related to one another that it is difficult to distinguish them apart. It has been shown from historical and geographical records¹ that the Mālpāhāriās separated from the parent stock between the years 1778 and 1819, and the cultural data also support the close relation between the two tribes. The view² that 'the Rajmahal Paharias, or as they are better known as Sauria or Mal Paharias, belong to a race distinct from the Mal Paharias of the south,' cannot be entertained at all, in the light of the results obtained in this paper. Further, the Sauria Paharias are known as the Mālér and not 'Mal Paharias,' as stated above. Risley pointed out this fact long ago, and his arguments have remained unnoticed both by the Government and by some recent writers. It would be interesting in this connection

¹ S. Sarkar, 'The Origin of the Mālpāhāriās,' *The Indian Historical Quarterly*, vol. ix, No. 4 (1933), p. 886.

² R. Ghosh, 'Note on the Sauria or Maler Paharias,' *Census of India*, Delhi, 1935, vol. i, Pt. III, B, p. 112.

to give prominence to the following paragraph from Risley : ¹

' Mr. Droese, with whom I discussed the subject some years ago, considered the two tribes to be of common origin. This view is borne out by the fact that they understand one another's language readily, though they assert that the dialects are different ; as well as by the identity of their ancestral worship, a point in which all races display a remarkably conservative spirit. As for the antagonism, which at present prevails between the north and south Paharias, it is only another instance of what has been commonly observed in Chota Nagpur, that two sections of the same tribe which in any way happen to become separated are often far more strongly opposed to one another than if they had been originally distinct tribes.'

The latter part of Risley's observations can be very well substantiated from the present state of the Mālér's and the Mālpāhāriās. The latter are found to bear a great dislike for the Mālér's, since they eat cows and dead animals.

In regard to Mālér-Orāon affinities, we find that appreciable differences exist between them in regard to stature, auricular height of head, orbitonasal breadth, orbitonasal arc, total facial length, sagittal arc of head, transverse arc of head, length height index, breadth height index, nasal elevation index, total facial index, and vertical cephalofacial index. That there is also some difference in the form of the head is evident from the large difference in the head height and in the length of the head ; this difference is as many as six times the difference between the Hill Mālér's and the Plains Mālér's and ten times that between the Hill Mālér's and the Mālpāhāriās. It has already been shown in what ways the Orāons differ characteristically from the Mālér's, and it would therefore be difficult to assume any close relationship between the two. The dissimilarity of the cultures of the two peoples also offers an additional proof of the difference between them. It thus appears that the Mālér's and the Orāons are two independent Dravidian-

¹ H. H. Risley, *The Tribes and Castes of Bengal*, vol. ii (Calcutta, 1892), pp. 66-72.

speaking peoples who never came into intimate contact with one another. The Mālér's may be taken as the autochthonous inhabitants of these hills, where they are still found to be living ; they undoubtedly appear to be one of the earliest remnants of the pre-Dravidian tribes inhabiting this particular region.

In conclusion, the author acknowledges his grateful thanks to Sir J. C. Bose, F.R.S., for his kind interest and encouragement. The author expresses his thanks to Dr. B. S. Guha, M.A., Ph.D., Anthropologist, Zoological Survey of India, for his guidance and valuable suggestions. My thanks are also due to the Government of Santal Perganas and to the officers of the Forest Department for the kind assistance which was offered to me in obtaining the data for these investigations.

XI.—THE ELECTRON-AFFINITY OF WATER

BY

ARUN KUMAR DUTTA, D.Sc.

IN interpreting the spectra of the alkali halide solutions, Franck and Scheibe¹ took into consideration the electron-affinity of water. With the alkali halide solutions they obtained an absorption at the extreme ultra-violet, which they ascribed to the removal of an electron from the halide ion. The equation connecting the energy amounts was put forth as

$$h\nu = E_x + H + P - S - E_{H_2O}$$

where ν is the frequency of the absorbed light,
 E_x is the electron-affinity of the halogen ion,
 H is the work of hydration of the halogen ion,
 P is the potential energy of the water dipole,
 S is the heat of dilution of the halogen atom,
and E_{H_2O} is the electron-affinity of water.

With the experimentally obtained value for ν , and introducing the other values of the thermochemical entities, they obtained 18.5 K. cal. as the electron-affinity of water.

Pauling² has later pointed out that the expression for the electron-affinity of water as obtained by Franck and Scheibe needs some modification. According to Pauling's contention, the previous equation of Franck and Scheibe should be modified in the form

$$E_x = h\nu + E_{H_2O} - 2H_x,$$

neglecting smaller quantities.

H_z in the above equation represents the contribution of the water dipoles to the heat of hydration of the halide ion. He has further pointed out that the value of ν used by Franck and Scheibe needed some alteration, as they had used the frequency corresponding to the maximum of absorption and not to the beginning of absorption, as is required in the calculation. However, putting in the values used by Franck and Scheibe, Pauling has obtained approximately 88 K. cal. for the electron-affinity of water.

To adduce support for the value for electron-affinity of water as calculated above, Pauling has tried to show that a rough estimate of the electron-affinity of water can be obtained from the value of specific magnetic susceptibility of water. He has pointed out that the average potential in a medium is given by

$$V_0 = \frac{2\pi eN}{3\nu} \sum \overline{r_k^2}$$

Introducing the expression for the molal diamagnetic susceptibility

$$\chi = - \frac{Ne^2}{6m_0c^2} \sum \overline{r_k^2}$$

one obtains

Replacing χ/ν by the specific magnetic susceptibility κ and introducing the numerical value of the constants,

$$V_0 = -6.43 \times 10^6 \kappa \text{ volts,}$$

or, considering the terms of potential energy of the electron W_0 , we can write

$$W_0 = -6.43 \times 10^6 \kappa \text{ volt-electrons.}$$

Pauling has argued that the electron-affinity E of a liquid or solid should be between $-W_0$ and $-1/2 W_0$, and putting in the value of the specific susceptibility of water he has

calculated the value of W_0 to be 4.63 e.v. The value of the electron-affinity of water calculated by Pauling certainly satisfies the criterion that it should be between $-W$ and $-1/2 W_0$.

But it may be pointed out here * that for calculating the electron-affinity of water, one should take into consideration the specific magnetic susceptibility of the water ion and not of the water molecules, as Pauling has done. This will involve a change in the value of $\sum_k \overline{r_k^2}$ in the previous equations and a consequent change in the estimate of the electron-affinity value.

Besides the region of absorption interpreted by Franck and Scheibe, alkali halide solutions generally show other patches of absorption. I undertook the investigations of the absorption spectra of the alkali halides in greater detail. The results of these investigations have been already reported.³ It had been shown there that, calculating on the lines of Pauling, the value for the electron-affinity of water was modified to some extent, the amount coming up to approximately 106 K. cal. A region of absorption patch with the alkali chloride solutions also corresponded to the same energy value. This patch of absorption had been repeatedly observed by various investigators and some of them tried to explain it as being due to impurities. From the correspondence of the position of the absorption patch and the electron-affinity value calculated, it was suggested by me that the absorption might be due to the electron-affinity of water. If any electron is bound up with the water molecules with such a large force, it is quite natural to expect an electron-affinity spectrum of the entity.

With a view to finding out other arrangements which might produce water ions and hence directly furnish other

* I am indebted to Dr. R. C. Mazumdar for kindly pointing out this defect in the arguments of Pauling.

evidence for the electron-affinity of water, I have now arranged for the production of sparks under water, using different metal electrodes and studying the absorption spectrum of water under varying conditions.

The experimental arrangement used was to pass condensed sparks under water. The chamber containing the water was covered in such a way that the light emanating from the spark did not find its way to the spectroscope. Two transverse quartz windows allowed the light from a hydrogen discharge tube to pass through the water and to be recorded in a small quartz spectrograph. Distilled water and pure metal electrodes were used in the experiments so far carried out. It was observed with copper electrodes that there was a short region of selective absorption in the region of λ 2600° corresponding to approximately 107 K. cal. This region of absorption was marked by general diffuse absorption extending towards longer wavelength, if the spark was allowed to run for a time longer than about 10 minutes. It has been sometimes observed with a shorter time of exposure that the region of selective absorption was not masked by the diffuse absorption. From the nature of the spectrum and the colour of the solution it could be generally inferred that the broad region of diffuse absorption was due to suspended material produced by the effect of spark.

A similar kind of spectrum was also observed with silver electrodes and with iron electrodes. But with electrodes of zinc, cadmium and bismuth, the selective absorption was not noticed. The broad diffuse absorption, however, appeared with a prolonged time of spark discharge.

The agreement between the energy corresponding to the ultra-violet selective absorption obtained by passing under-water spark with copper electrodes and the electron-affinity of water is quite good, and one is tempted to say that by the process of under-water spark some water molecules have electrons attached to them.

With a view to finding out another possible origin of this selective absorption, the solution obtained after passing the spark through copper electrodes in water for about 15 minutes was ultra-filtered.* An absorption spectrum of the resulting ultra-filtered solution showed that the general diffuse absorption was no longer observed, but the selective absorption was persistent. When the solution is kept in a stoppered flask, it appears that with the lapse of time the intensity of its absorption spectrum gradually fades.

In order to test for the presence of any copper compound produced by the electrode discharge and dissolved in the solution, spot test for copper was carried out with α Benzoin-Oxime.⁴ This method gives a positive sign for the copper radical when present to the limit of 10^{-7} grm. in a drop. With the ultra-filtered solution, however, this gave a negative test for copper. The above procedure generally eliminates the possibility that any copper salt dissolved in the solution gives the selective absorption spectrum. To make the point sure, comparison spectra were taken with different copper salt solutions prepared in the laboratory. Comparatively stronger solutions of copper sulphate, copper chloride and copper acetate were prepared and the absorption spectra through them were photographed to indicate the nature of absorption with these salts. Very weak solutions of these salts were prepared by dissolving 0.2 grm. of these substances in 5000 c.c. of distilled water. These solutions gave the above-mentioned spot tests quite well, but did not show any absorption spectrum in the quartz region. This precludes any possibility of a copper salt giving the spectrum at λ 2600° approximately.

Thus there seems to be a possibility that the electron-affinity of water might have caused the above-mentioned spectrum. But more thorough and critical examination of the hypothesis should be made, before one makes any

* I am indebted to Prof. J. N. Mukherjee, Khairat Professor of Chemistry, Science College, Calcutta, for kindly arranging to ultra-filtrate the solutions for me.

definite assertion about it. A more thorough investigation will be carried out in future.

I beg to express my heartfelt thanks to Sir J. C. Bose, F.R.S., for the facilities offered at the Bose Research Institute, and to Prof. D. M. Bose, Ph.D., for helpful discussions.

REFERENCES

- ¹ FRANCK and SCHEIBE : *Zeits. f. Phys. Chem.*, vol. 139 (1928), p. 22.
 - ² PAULING : *Phys. Rev.*, vol. 34 (1929), p. 954.
 - ³ DUTTA : *Trans. Bose Res. Inst.*, vol. x.
 - ⁴ FEIGGEL : *Ber.*, vol. 56 (1923), p. 2083 ; *Microchem.*, vol. 1 (1923), p. 76.
- AZZALIN : *Ann. Chim. Appl.*, vol. 15 (1925), p. 373 (B. D. Reagents).

XII.—ON THE ANGULAR DISTRIBUTION OF COSMIC RAYS

BY

RADHESH CHANDRA GHOSH, PH.D. (TÜBINGEN)

INTRODUCTION

(a) *Discovery of cosmic rays.*—In 1912 Hess¹ observed that the ionisation in a closed ionisation chamber increases as one measures it in higher and higher altitudes. To account for this phenomenon he propounded the hypothesis that this ionisation was due to certain kinds of rays, and he called them ultra-rays. Kolhörster² in Germany and Millikan³ in America afterwards repeated this experiment. Millikan at first thought that Hess's observation was spurious; but afterwards with improved technique of measurements he confirmed the existence of these rays. Millikan at the same time proposed the theory that these rays consist of high-energy photons and named them cosmic rays. Piccard⁴ and, more thoroughly, Regener⁵ measured the intensity of these rays in higher altitudes. They all supported Hess's observation.

(b) *The nature of radiation.*—That the cosmic radiation consisted in part at least of high-energy, electrically charged particles was shown by the investigation of Clay,⁶ who found a variation in the intensity of these rays in different magnetic latitudes. This was attributed to the action of the earth's magnetic field in deflecting a part of the charged particles which were contained in the original beam. Afterwards Compton⁷ and his fellow workers made an elaborate

study of this geomagnetic effect and confirmed Clay's observation. Compton showed that these particles in order to penetrate the earth's atmosphere must possess an energy of the order of $10^{10}e$ volt, and were possibly electrically charged particles. The rays which reach the equator of the earth, if they be electrically charged particles, should possess an energy greater than $4 \cdot 10^{10}e$ volt, or they should be photons and neutrons. Bothe and Kolhörster,⁸ by their coincidence experiment, detected the presence of high-energy, electrically charged particles.

(c) *East-west asymmetry*.—If cosmic rays be electrically charged particles, the earth being a magnet should have an effect on these particles according to the sign of the charge. If they be positively charged they will be deviated from the west to the east, whereas the opposite will be the case if they be negatively charged particles. If one can measure the intensity from the east as well as from the west directions (azimuthal angles), such measurements should give an insight into the nature of these rays. Various experiments were performed at different places, and it was found that the positively charged particles always predominate over the negative ones, and the percentage of predominancy of positively charged particles increases with higher altitudes and lower geomagnetic latitudes.

(d) *North-south asymmetry*.—Recent experiments by the method of multiple coincidences carried out by Johnson⁹ in Mexico (geomagnetic latitude 29° N.) and by Clay¹⁰ in Java (geomagnetic latitude 18° S.) have shown that in the northern geomagnetic hemisphere the intensity of cosmic rays in geomagnetic meridian is, for equal zenith angles, greater from the south than from the north; conversely, in the southern hemisphere it is greater from the north than from the south. That this is a consequence of the earth's magnetic field on the motion of the charged particles was pointed out by Lemaitre and Vallatra¹¹ as early as 1932. Shortly afterwards Bouckaert¹² was able to calculate this north-

south asymmetry for geomagnetic latitudes up to 20° and moderate zenith angles. Starting from the assumption of an isotropic distribution of charged particles at very great distance from the earth, it was found that, due to the influence of the earth's magnetic field, some of these particles are shot back to infinity, while others are allowed to reach the earth. It is shown that at any point, at a given distance from the earth's magnetic centre, all particles of a given energy reaching the point must come within a cone generally of very involved shape, which forms the boundary between the region where all directions are allowed and the region in which only some or no directions are allowed. With the help of very elaborate analysis the results obtained by Lemaitre and Vallatra have been embodied in the form of curves, drawn for different magnetic latitudes, which give the least energy required of a particle (expressed in units of milliströmer) to arrive at any zenith angle in the meridian plane, at any point on the earth up to latitude 40° . It is found that these values are not the same for the same north and south zenith angles corresponding to a given magnetic latitude, and hence a north-south asymmetry arises. This asymmetry is irrespective of the sign of the particles. It is found that the minimum of the north-south asymmetry found by Johnson at geomagnetic latitude 29° N. at about 45° zenith angle, as well as the general features of these experimental results, are fully accounted for by the earth's magnetic field. According to these authors further experimental study of this asymmetry will provide a direct workable method for the analysis of spectral distribution of corpuscular cosmic radiation independent of its sign.

(e) *Cosmic ray showers and bursts.*—The interactions of penetrating cosmic radiation with matter have given rise to some very interesting results, *i.e.* the production of showers and the so-called Hoffman's bursts. From the cloud chamber photographs of these showers in a magnetic field Anderson discovered the existence of positron—a

positively charged particle of the same mass as that of electron.

Geiger and Funfer¹³ tried to give a systematic account of the cosmic rays by the assumption that cosmic rays are high-energy, positively charged particles which are successively converted into photons, electron pairs, etc. This, however, cannot satisfactorily account for the presence of showers. Heisenberg¹⁴ has extended Fermi's theory of β ray disintegration to give a quantitative picture of shower production.

There has been no systematic study of the cosmic rays in this part of India, and it is proposed to undertake a series of investigations on the different aspects of cosmic ray activities which have been summarised above. The Bose Research Institute has two research stations, one at Calcutta (mag. lat. 23° , altitude 70 ft.) and another at Darjeeling (mag. lat. 27° N., altitude 7100 ft.). In the present paper an account is given of the study of the two problems of east-west and north-south asymmetry at these two places. Other investigations are in hand and will be reported in subsequent publications.

EXPERIMENTAL ARRANGEMENTS

The experiment was carried out with three Geiger-Müller counters by the method of coincidence.

The counter consisted of a brass tube with a central wire fixed to two ebonite pieces at its ends. This central wire was of steel of 0.2 mm. diameter and was cleaned with sandpaper and benzene and then oxidised by passing a momentary current of 2 amperes. The counters had an air pressure of 6 cm. Hg. If the diameter of ebonite pieces at the ends of the counters be equal to the diameter of the inner brass tube as shown in fig. 56, the counters do not work satisfactorily during hot summer months. For these new counters were prepared as shown in fig. 57. In this type of counter the electric field near the ebonite is made

relatively more divergent. With counters of the type 1 the counting voltage, *i.e.* the region of voltage over which the number of counts per unit of time practically remains constant, is about 50 volts, while that with the second type of counters is about 80 volts.

A rotating bench was prepared in which two or three counters could be placed one above the other and the whole system could be rotated up to an angle of 90° against the vertical in any direction. When the east-west intensity was measured, the rotating bench, and along with it the counters, were placed along the magnetic meridian, and by

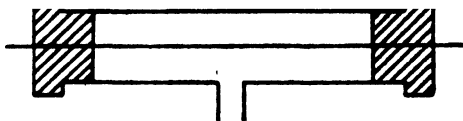


FIG. 56. Type 1.

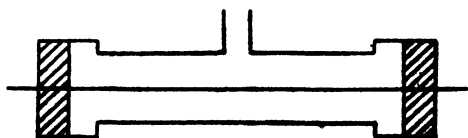


FIG. 57. Type 2.

suitably tilting the plane of the counters, the intensity of the rays coming along any azimuthal angle could be measured.

The case was similar, of course with reverse arrangement, when we measured the intensity in zenith angles.

The amplifier was of the type used by Bothe¹⁸; in the case of two-tube coincidence five valves were used—one of them was a screen grid valve, A 442, and the last one was a power valve. In the case of three-tube coincidence seven valves were used—two of them were screen grid ones. All the grid voltages, condensers, resistances, as well as anode voltages, were adjusted by trial. With the power valve instead of

a loud-speaker or a high-frequency oscillograph, an electromagnetic recorder similar to that of a telephone call counter was used.

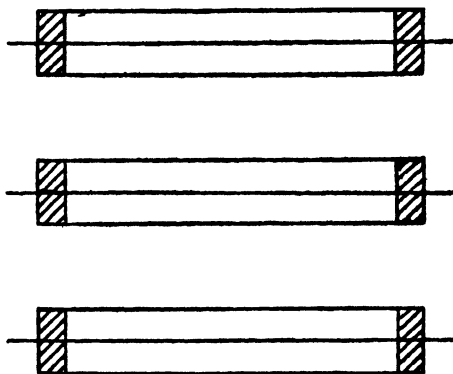


FIG. 58.

RESULTS OF THE INTENSITY IN DIFFERENT AZIMUTHAL ANGLES

This experiment at Calcutta was carried out with counters as shown in fig. 58, and the readings were naturally taken in winter.

All other readings given in this paper were taken with the second type of counters and with the geometry as given in Table I.

TABLE I.—GEOMETRY OF MEASUREMENT

Length of counter	Diameter of the counter	Distance between the counters
Type 1. 30 cm. .	3.0 cm.	3 cm.
Type 2. 25 cm. .	2.5 cm.	5 cm.

In making a set of measurements, first the vertical intensity is measured, then the frame is turned to a given

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azimuthal angle, *i.e.* 30° east, and the reading is taken. Next the frame is turned to the azimuthal angle to the west, *i.e.* 30° west, and the reading is taken. By taking the readings for equal azimuthal angles in succession, we reduce to a minimum any variation in readings which might arise owing to the variation in time of the constants of the apparatus, *i.e.* the gas pressure in the counter tubes and the applied voltage.

Over and above, in this method one does not require any extraordinarily long time constancy of the apparatus. After each series of experiments a check reading was always taken for the vertical intensity. When the same experiment was repeated the vertical intensity again provided us with a check reading. In case of any unusual counting in the middle of a series of experiments, the whole series was rejected and a new series was then begun. For each position at least 100 counts were taken. This gave us a 10 per cent. statistical error for each position. But in drawing a curve, since all the positive errors could not occur in one direction while the negatives in the other, the values obtained from the mean curve are on the whole much more accurate than the individual readings.

RESULTS AT CALCUTTA

Latitude 23° , altitude 70 ft. Results of the east-west intensity distribution with two Geiger-Müller counter coincidences :

Angles against the vertical							Number of counts per minute
0°	4.44
30° west	3.89
30° east	3.07
45° west	2.91
45° east	2.18
60° west	1.77
60° east	1.47

Results of the east-west intensity at Calcutta with three Geiger-Müller counters :

Experiment 1. Type 1

Angles against the vertical	Number of counts per minute
0°	2.86
30° west	2.15
30° east	1.67
60° west	1.17
60° east	0.97

Experiment 2. Type 1

Angles against the vertical	Number of counts per minute
0°	2.94
30° west	2.18
30° east	1.79
60° west	1.15
60° east	0.97

Experiment 3. Type 1

Angles against the vertical	Number of counts per minute
0°	2.87
30° west	1.94
30° east	1.89
60° west	1.23
60° east	1.00

Mean results of these three sets of readings :

Angles against the vertical	Number of counts per minute
0°	2.89
30° west	2.06
30° east	1.78
60° west	1.18
60° east	0.98

RESULTS OF EAST-WEST INTENSITY AT DARJEELING

Latitude 27° , altitude 7100 ft.*Experiment 1. Type 2*

Angles against the vertical	Number of counts per minute
0°	2.54
30° west	2.00
30° east	1.53
45° west	1.02
45° east	0.86
60° west	0.74
60° east	0.56

Experiment 2. Type 2

Angles against the vertical	Number of counts per minute
0°	2.48
30° west	1.81
30° east	1.36
45° west	1.06
45° east	0.82
60° west	0.76
60° east	0.52

Experiment 3. Type 2

Angles against the vertical	Number of counts per minute
0°	2.61
30° west	1.82
30° east	1.45
60° west	0.76
60° east	0.45

Mean results of these three sets of readings :

Angles against the vertical	Number of counts per minute
0°	2.54
30° west	1.88
30° east	1.45
45° west	1.04
45° east	0.84
60° west	0.75
60° east	0.51

A COMPARATIVE STUDY WITH OTHER WORKS ON EAST-WEST INTENSITY DISTRIBUTION OF COSMIC RAYS

Names of workers	Geomagnetic latitude	Altitude in meters	Angle against vertical	West-east predominancy in per cent.
Johnson ⁽¹⁶⁾	57°	1900	20° 30° 40°	Just above the experimental error
Johnson ⁽¹⁷⁾	29°	2250	30° 35° 45° 50° 55° 65°	6.8 ± 1.9 6.3 ± 1.3 10.2 ± 1.7 11.5 ± 1.8 10.3 ± 1.8 5.5 ± 2.7
Alvarez ⁽¹⁸⁾ and Compton	29°	2310	15° 30° 45°	3.0 ± 2.0 5.5 ± 0.8 2.5 ± 1.0
Johnson ⁽¹⁹⁾ and Stevenson	51°	0	15° 30° 45°	1.0 ± 0.7 3.2 ± 0.8 2.5 ± 1.0
Johnson ⁽²⁰⁾	0°	0 3500 and 4500	15° 30° 45° 60°	0 m. 3500 m. 4500 m. 6 6.5 7 8 10 13 10 13 14 — 12 13
Stevenson ⁽²¹⁾	48°	3100	30° 45°	2.2 ± 0.6 2.3 ± 0.7
Rossi ⁽²²⁾	11°	2370	15° 30° 45°	11.0 ± 1.8 15.7 ± 1.8 18.8 ± 2.2
Messer-schmidt ⁽²³⁾	52°	33	25° 45° 55°	0.18 ± 0.02 0.29 ± 0.02 0.14 ± 0.02
Results of this exp. at Calcutta	23°	20	30° 60°	12.3 ± 1.4 18.6 ± 2.0
Results of this exp. at Darjeeling	27°	2150	30° 45° 60°	25.8 ± 3.0 21.3 ± 2.0 31.6 ± 3.5

As Leprince Ringuet and Auger ²⁴ have published only the curves and not the data, it was not possible to calculate the percentage. But their curves are similar to those that are found in this experiment.

Although the readings at Calcutta and at Darjeeling are taken with two different sets of counters having varied

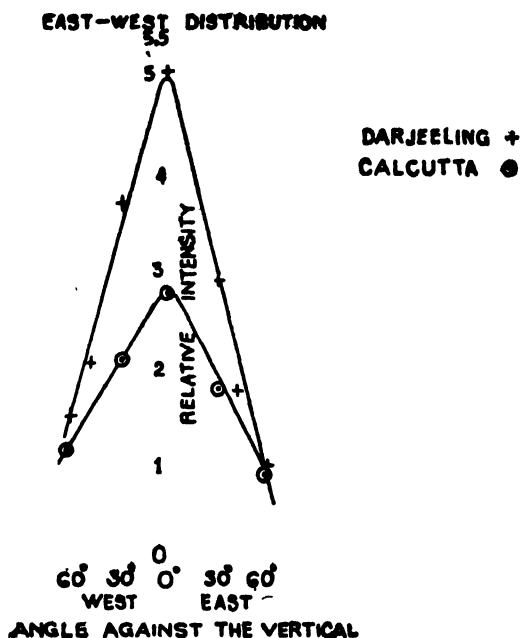


FIG. 59.

dimensions, and form, as well as the distance between them, thereby the angular definition, nevertheless it is interesting to put them in a simple curve and to have a direct comparison between them.

As the intensity along 60° east is a minimum, the number of counts for this angle at Darjeeling is multiplied by 2 to make it coincide with that at Calcutta. All other readings at Darjeeling are correspondingly doubled. We see from the curves so drawn (fig. 59) that the percentage of

predominancy of the positively charged particles at Darjeeling, *i.e.* at a higher altitude, is greater than that at Calcutta. The ratio of the 60° east to the vertical intensity at Calcutta is about 3, while that at Darjeeling is about 5. This, however, depends on the angular definition and also on the altitude and the azimuthal angle. The difference in the readings of the counters at these places is proportional to the absorption due to a column of air of thickness $L \cos \theta$, where L is the difference in elevation and θ is the azimuthal angle. On the east of the room in which this experiment was carried out at Darjeeling, there is a small hill which may slightly affect the result.

The result of this experiment at Calcutta agrees quite well with that of Rossi. It does not, however, agree so well with other authors, which probably arises from the difference in geometry. The other authors did not publish any statement about their arrangement in their paper, so a comparison of geometry was not possible.

READING OF THE NORTH-SOUTH INTENSITY AT CALCUTTA AND THAT AT DARJEELING

This series of experiments was carried out in the summer with the second type of counters. The same rotating bench was used.

Results of the north-south intensity at Calcutta with two Geiger-Müller counters :

Angles against the vertical	Number of counts per minute
0°	2.87
15° south	2.37
15° north	2.04
30° south	2.08
30° north	1.63
45° south	1.68
45° north	1.33
60° south	1.03
60° north	0.77

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Results of the north-south intensity at Calcutta with three Geiger-Müller counters :

Experiment 1. Type 2

Angles against the vertical						Number of counts per minute
0°	1.16
30° south	0.86
30° north	0.65
45° south	0.68
45° north	0.48
60° south	0.44
60° north	0.33

Experiment 2. Type 2

Angles against the vertical						Number of counts per minute
0°	1.23
30° south	0.91
30° north	0.66
45° south	0.63
45° north	0.45
60° south	0.39
60° north	0.33

Mean results of the last two sets of readings :

Angles against the vertical						Number of counts per minute
0°	1.19
30° south	0.88
30° north	0.65
45° south	0.65
45° north	0.46
60° south	0.42
60° north	0.33

Results of the north-south intensity at Darjeeling with three Geiger-Müller counters :

Experiment 1. Type 2

Angles against the vertical						Number of counts per minute
0°	2.53
30° south	1.99
30° north	1.68
60° south	0.74
60° north	0.52

Experiment 2. Type 2

Angles against the vertical	Number of counts per minute
0°	2.71
30° south	1.84
30° north	1.43
60° south	0.76
60° north	0.55

NORTH-SOUTH INTENSITY DISTRIBUTION

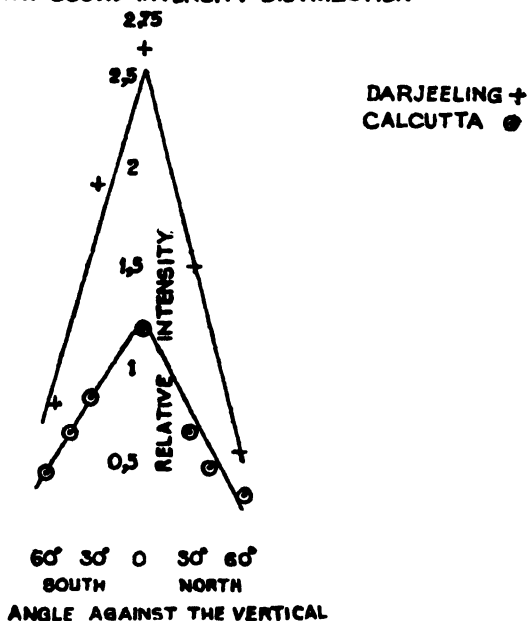


FIG. 60.

Experiment 3. Type 2

Angles against the vertical	Number of counts per minute
0°	2.48
30° south	1.95
30° north	1.39
60° south	0.84
60° north	0.61

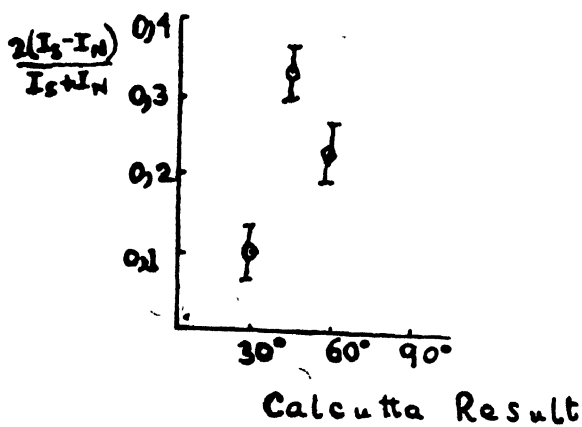
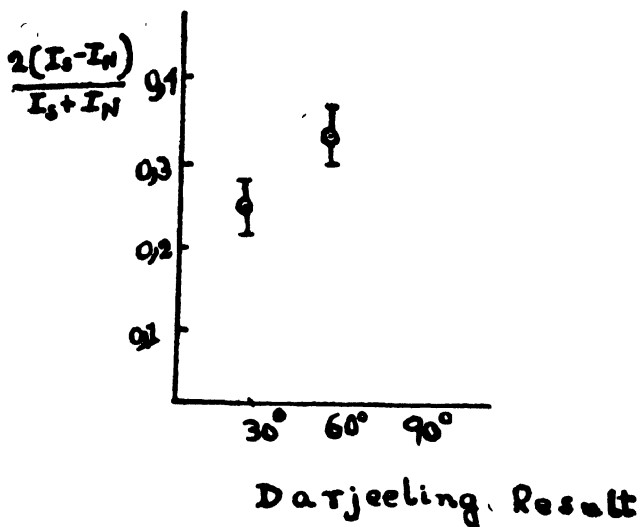


FIG. 61.

Mean results of the three sets of readings which were taken at Darjeeling :

Angles against the vertical	Number of counts per minute
0°	2.57
30° south	1.93
30° north	1.50
60° south	0.78
60° north	0.56

As the readings of the north-south intensity distribution at Calcutta, as well as at Darjeeling, were taken with the same counters and geometry, we get a direct comparison from the figure.

CONCLUSION

(1) The intensity of the cosmic rays from the west predominates over that from the east at both the stations. But this predominancy is much more prominent at Darjeeling than at Calcutta. This shows that the positively charged particles are found more than the negative ones in cosmic rays, and this effect increases with the altitude.

(2) The intensity of the cosmic rays from the southern directions in this hemisphere predominates over that from the northern direction. This effect also increases with the altitude. But a point of dip in the curve, *i.e.* at a certain angle at which the northern intensity of cosmic rays equalises with that of the southern intensity, as observed by Johnson, could not be observed in the case of the angles measured (see fig. 61).

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REFERENCES

- ¹ V. F. HESS : *Phys. Zs.*, vol. 13 (1912), p. 1084 ; *Wiener Ber.* (2A), vol. 121 (1912), p. 2001.
- ² W. KOLHÖRSTER : *Phys. Zs.*, vol. 14 (1913), pp. 1066, 1153 ; *Abh. Naturforsch. Ges. Halle N.F.* (1914), No. 4 ; *Verh. Dtsch. Phys. Ges.*, vol. 16 (1914), p. 719 ; *Berl. Ber.*, vol. 34 (1923), p. 366.
- ³ R. A. MILLIKAN : *Phys. Rev.*, vol. 23 (1924), p. 778 ; *Nature* (London), vol. 116 (1925), p. 823.
- ⁴ A. PICCARD and M. COSYONS, *C.R.*, vol. 195 (1932), p. 604.
- ⁵ E. REGENER : *Nature*, vol. 130 (1932), p. 364 ; *Naturwissenschaften*, vol. 20 (1932), p. 695 ; *Phys. Zs.*, vol. 34 (1933), p. 306.
- ⁶ J. CLAY : *Proc. Roy. Acad.* (Amsterdam), vol. 30 (1927) p. 1115 ; vol. 31 (1928), p. 1091.
- ⁷ A. M. COMPTON : *Phys. Rev.*, vol. 43 (1933), p. 387 ; *Nature*, vol. 131, (1933), p. 713.
- ⁸ W. BOTHE and W. KOLHÖRSTER : *Zs. f. Phys.*, vol. 56 (1929), p. 751 ; *Phys. Zs.*, vol. 30 (1929), p. 516.
- ⁹ T. H. JOHNSON : *Phys. Rev.*, vol. 47 (1935), p. 91.
- ¹⁰ J. CLAY : *Physica*, vol. 8 (1935), p. 867.
- ¹¹ G. LEMAITRE and M. S. VALLATRA : *Phys. Rev.*, vol. 42 (1932), p. 914 ; vol. 43 (1933), p. 87 ; vol. 49 (1936), p. 719.
- ¹² L. BOUCKAERT : *Ann. de la Soc. Sci. de Bruxelles*, vol. A54 (1934), p. 174.
- ¹³ H. GEIGER and E. FUNFER : *Zs. f. Phys.*, vol. 93 (1934-35), p. 543.
- ¹⁴ W. HEISENBERG : *Zs. f. Phys.*, vol. 101 (1936), p. 533.
- ¹⁵ W. BOTHE : *Zs. f. Phys.*, vol. 59 (1929), p. 1.
- ¹⁶ TH. H. JOHNSON : *J. Franklin Institute*, vol. 214 (1932), pp. 665, 689.
- ¹⁷ TH. H. JOHNSON : *Phys. Rev.*, vol. 43 (1933), p. 381.
- ¹⁸ L. ALVAREZ and A. H. COMPTON : *Phys. Rev.*, vol. 43 (1933), p. 835.
- ¹⁹ TH. H. JOHNSON and E. C. STEVENSON : *Phys. Rev.*, vol. 44 (1933), p. 125.
- ²⁰ TH. H. JOHNSON : *Phys. Rev.*, vol. 44 (1933), p. 856.
- ²¹ E. C. STEVENSON : *Phys. Rev.*, vol. 44 (1933), p. 855.
- ²² B. ROSSI : *Phys. Rev.*, vol. 45 (1934), p. 212.
- ²³ W. MESSERSCHMIDT : *Zs. f. Phys.*, vol. 87 (1934), p. 800.
- ²⁴ LEPRINCE RINGUET and AUGER : *Nature*, vol. 133 (1934), p. 138.

XIII.—THE THEORY OF PROPAGATION OF RADIO-WAVES IN UPPER ATMOSPHERE

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(Premchand Roychand Scholar)

ABSTRACT

THE present paper offers a generalised theoretical investigation of the Theory of Propagation of Radio-Waves in Upper Atmosphere, and presents the formulation of a general expression for dispersion and absorption, which includes the Lorentz polarisation correction as well. The mathematical treatment differs considerably from the old and traditional method as was previously adopted in ionospheric investigations and follows closely the lines of thought and principles of new quantum mechanics. The formulae, developed in the present paper, reduce to those recently obtained by the author¹ when the polarisation correction is neglected. It is shown that the well-known Appleton-Hartree formula, which is so extensively used in all ionospheric investigations, comes out only as a particular case.

INTRODUCTION

In a recent paper* communicated to *Zeitschrift für Physik*¹ a new mathematical method of attack is developed to investigate the theory of dispersion and absorption of radiowaves by upper atmospheres. The new method is an

* This paper will be referred to as MI.

outcome of direct application of the principles and conceptions of new quantum mechanics to the problem of ionosphere and differs considerably from the old and traditional representation of the theory worked out by Appleton,² Hartree,³ Goldstein⁴ and others. The Appleton-Hartree formula, as shown, is, so to say, qualitative in nature, lacking in rigour and detail, and is true in its present form under *ad hoc* assumptions only.

Although the theory, previously developed in MI, correlated a number of interesting experimental facts and proved very successful in investigating the structure of ionosphere, we, however, left the question open as regards the nature of the forces determining the motion of electrons. We simply took the space average value of the electric field and neglected the Lorentz polarisation correction. But it is well known that during the investigation of the alteration of dielectric constant due to the presence of ordinary neutral particles between condenser plates, the Lorentz polarisation correction, which takes account of the actual corpuscular distribution of matter, must be added to the usual 'pipe force'; the resulting mathematical formula for dielectric constant being tested experimentally and found to be in good agreement. Since the ionosphere contains electrons in presence of a large number of positive ions and neutral particles, Hartree advocated the introduction of this Lorentz correction and included it in his theoretical investigation of the formula for complex refractive index. The resulting Appleton-Hartree formula has been since then thoroughly studied and applied extensively in all ionospheric investigations.⁵ In 1933 Tonks⁶ raised the question on the validity of Hartree's inclusion of the polarisation term and emphasised along with Norton⁷ that the correction term should be omitted except where 'there is some detailed arrangement of the negative with respect to the positive charges.' Hartree⁸ had afterwards pointed out some unjustified assumptions in the arguments put forward by Tonks, which

put the matter again in great confusion. These discussions, however, led Darwin⁹ to take up the subject and examine it thoroughly from all possible angles. He attacked the problem by an entirely new method which avoided the ambiguities involved in the analysis of internal electric fields in matter, in which we visualise the individual atom or molecule as an object enclosed in a spherical cavity, influenced by the field due to induced charges on the walls. This analysis of Darwin has shown that the conditions which prevail in ionosphere and in metals would favour the omission of the Lorentz polarisation correction.

It is, however, to be frankly admitted that the discussion on the subject is very complicated, and any conclusion should therefore be accepted with caution. In the present investigation we shall try to find a solution of the problem in an indirect way, and shall therefore refrain from any direct discussions on the correction term due to polarisation. We shall rather accept it as it is and attempt to generalise our previous formulae for dispersion and absorption so as to include the Lorentz polarisation correction. Attempt will then be made to obtain any clue, if possible, from a comparison of these theoretical results with those obtained from the wireless observations, with a view towards a solution of this controversy regarding the nature of the accelerating field on the motion of the electron.

§ 1. Let us consider the ionosphere as consisting of particles (ionised and neutral atoms and molecules) which are distributed at random and electrons which are moving in the field of force of the particles. We shall thereby assume the heavy particles as a massive centre of force, the collisions between electrons and particles taking place elastically. Our object is now to investigate the change in the distribution function of the electrons in such a system, subject to a simply periodic field of the incident plane electromagnetic field travelling along the direction of the axis of z and a constant magnetic field due to the earth, resolved into two

components, H_z along the axis of z and H_x perpendicular to it. The resulting change in the distribution function f of the electron due to the combined action of the electric and magnetic field on the one hand and the collisions on the other hand is given by

$$\frac{\partial f}{\partial t} + \left(\vec{v} \text{ grad } f \right) + \left(\text{grad } f, \frac{\vec{F}}{h} \right)_{\vec{K}} + \iiint w(\vec{K}\vec{K}') \left[f(\vec{K}) - f(\vec{K}') \right] dK_x dK_y dK_z = 0 \quad (1)$$

where the second and the third terms are the change in the distribution function due to the electric and magnetic field and the last term is due to that of the collisions of the electrons with the particles. We shall denote it by

$-\left(\frac{\partial f}{\partial t}\right)_{\text{collision}}$ for future reference. $w(\vec{K}\vec{K}')$ is the transition probability of the electron from state \vec{K} to \vec{K}' , \vec{K} and

\vec{K}' being the initial and final wave vectors of the electron and \vec{v} its group velocity. \vec{F} is the Lorentz force given by

$$\vec{F} = e\vec{E} + \frac{e}{c}[\vec{v}\vec{H}] \quad (2)$$

where the intensity of the electric field

$$\vec{E} = \vec{E}_0 e^{i\omega t} \quad (3)$$

$\frac{\omega}{2\pi}$ being the frequency of the wave and \vec{H} the resultant of the external magnetic field. (The notations and symbols are the same as in previous paper MI. For convenience they are again given in an appendix at the end of this paper.)

We are now to solve the integro-differential equation (1), in which we substitute

$$f = f_0 + f_1, \quad f_1 \ll f_0 \quad (4)$$

For f_1 we make further substitution

$$f_1 = e^{i\omega t} (\vec{K} \vec{\chi}(\vec{K})) \quad (5)$$

where $\vec{\chi}(\vec{K})$ depends only on the magnitude of \vec{K} . By introducing the time of relaxation τ defined by

$$\left(\frac{\partial f}{\partial t} \right)_{\text{collision}} = -f_1/\tau \quad (6)$$

we obtain, after some simplifications with the help of the above equations, the expression for $\vec{\chi}$ as given by

$$\vec{\chi} = - \frac{U\vec{V} - [\vec{W}\vec{V}] + \frac{\vec{W}}{U}(\vec{W}\vec{V})}{U^2 + (\vec{W}\vec{W})} \quad (7)$$

When the medium is homogeneous and there is no temperature gradient we have

$$U = 1 + i\nu\tau, \quad \vec{V} = \tau \frac{h}{m} \frac{\partial f_0}{\partial \vec{z}} e\vec{E} \quad (8)$$

and

$$\vec{W} = \frac{\tau e}{mc} \vec{H}$$

Now following the usual method we obtain the expression for current given by

$$I = 2e \iiint \vec{v} f_1 dK_x dK_y dK_z \\ = S' E_0 \quad (9)$$

where the tensor S' stands for *

$$S' = K^t U^t - L^t \cdot [U^t \vec{w}_L]^t + M^t \cdot \vec{w}_L \vec{w}_L \quad (10)$$

$\vec{w}_L = \frac{e}{mc} \vec{H}$, which is double the Larmor frequency, and

* Explanation of symbols: $\vec{A}\vec{B}$, $(\vec{A} \vec{B})$ and $[\vec{A} \vec{B}]$ represent dyadic, scalar and vector products of two vectors \vec{A} and \vec{B} . The suffix 't' above means that the symbol in question is a tensor.

U^i is a unit tensor ; the tensors K^i , L^i and M^i , which are of diagonal nature, are given by

$$K^i = -\frac{2e^2\hbar^2}{m^2} \iiint \frac{\tau \vec{K} \vec{K} \frac{\partial f_0}{\partial \epsilon} (1 + i\omega\tau)}{(1 + i\omega\tau)^2 + \tau^2 \left(\frac{\vec{w}}{w_L} \cdot \frac{\vec{w}}{w_L} \right)} dK_x dK_y dK_z \quad (11)$$

$$L^i = -\frac{2e^2\hbar^2}{m^2} \iiint \frac{\tau^2 \vec{K} \vec{K} \frac{\partial f_0}{\partial \epsilon}}{(1 + i\omega\tau)^2 + \tau^2 \left(\frac{\vec{w}}{w_L} \cdot \frac{\vec{w}}{w_L} \right)} dK_x dK_y dK_z \quad (12)$$

$$M^i = -\frac{2e^2\hbar^2}{m^2} \iiint \frac{\tau^3 \vec{K} \vec{K} \frac{\partial f_0}{\partial \epsilon}}{(1 + i\omega\tau)^2 + \tau^2 \left(\frac{\vec{w}}{w_L} \cdot \frac{\vec{w}}{w_L} \right)} \frac{dK_x dK_y dK_z}{(1 + i\omega\tau)} \quad (13)$$

The components of $[U^i_{w_L}]^j$ are given by the scheme

$$[U^i_{w_L}]_{\alpha\beta} = \alpha = \begin{matrix} & \beta = x & y & z \\ \begin{matrix} x \\ y \\ z \end{matrix} & \begin{pmatrix} 0 & -w_x & 0 \\ w_x & 0 & -w_y \\ 0 & w_x & 0 \end{pmatrix} \end{matrix} \quad (14)$$

Having thus obtained the tensor S^i which connects the electric current with the field, we can now easily deduce the expressions for dispersion and absorption with the help of Maxwell's electrodynamics. Following Hartree we shall take the electric field \vec{E} at a point P in the medium (the field actually acting on the scattering element there) to be composed of

$$\vec{E} = \vec{E}^{(0)} - \vec{E}' + \vec{E}'' + \vec{E}^{(0)} \quad (15)$$

i.e. the electric field at P, say, is calculated by finding first the field $\vec{E}^{(0)}$ due to the smoothed continuous distribution over all space, subtracting the contribution \vec{E}' arising from the continuous distribution in a small sphere round P and adding the contribution \vec{E}'' due to the actual discrete

distribution inside the small sphere; $\vec{E}^{(i)}$ being the electric field of the incident wave. Writing

$$-\vec{E}' + \vec{E}'' = \beta \sigma' \vec{E} \quad . \quad . \quad (16)$$

where β is not likely to be far from $\frac{1}{2}$ and the scattering tensor σ' is given by

$$\sigma' = \frac{4\pi}{i\omega} S' = K' \cdot U' - L' \cdot \left[U' \vec{w}_L \right]' + M' \cdot \vec{w}_L \vec{w}_L \quad . \quad (17)$$

where

$$K' = \frac{4\pi}{i\omega} K', \quad L' = \frac{4\pi}{i\omega} L', \quad M' = \frac{4\pi}{i\omega} M' \quad . \quad . \quad (18)$$

We obtain thus from (15)

$$(U' - \beta \sigma') \vec{E} = \vec{E}^{(i)} + \vec{E}^{(s)} \quad . \quad . \quad (19)$$

For simplicity in the final formula let us now introduce two new vectors \vec{L} and \vec{D} , defined by

$$\vec{L} = (U' - \beta \sigma') \vec{E} \quad . \quad . \quad (20)$$

$$\vec{D} = \{U' + (1 - \beta) \sigma'\} \vec{E} \quad . \quad . \quad (21)$$

or eliminating \vec{E} from these we obtain

$$\vec{D} = \epsilon' \vec{L} \quad . \quad . \quad (22)$$

where

$$\epsilon' = U' + \sigma' (U' - \beta \sigma')^{-1} \quad . \quad . \quad (23)$$

or we can write it in a more convenient form :

$$\beta (\epsilon' - U') = (U' - \beta \sigma')^{-1} - U' \quad . \quad (24)$$

But on the other hand \vec{D} is related to \vec{L} by

$$\vec{D} = (n^2 \vec{U} - \vec{n} \vec{n}) \vec{L} \quad . \quad . \quad (25)$$

so that from (22) and (25)

$$(\epsilon' - n^2 U' + \vec{n} \vec{n}) \vec{L} = 0 \quad . \quad . \quad (26)$$

since $\vec{L} \neq 0$, the determinant of the components of $\epsilon' - n^2 U' + \vec{n} \vec{n}$, must be zero, and this gives an equation for the

evaluation of the refractive index which is evidently complex; the real part of this gives the dispersion and the imaginary one the absorption.

Now to find ϵ' we require the tensor reciprocal to $U - \beta\sigma$, which is given by

$$U - \beta\sigma = U - \beta K \left\{ U - \frac{L}{K} \left[U \vec{w}_L \right] + \frac{M}{K} \vec{w}_L \cdot \vec{w}_L \right\} \\ = (1 - \beta K) \left\{ U + \gamma L \left[U \vec{w}_L \right] - \gamma M \vec{w}_L \cdot \vec{w}_L \right\} \quad (27)$$

$$v = \frac{\beta}{1 - \beta K} \quad . \quad . \quad . \quad . \quad . \quad . \quad (28)$$

(For convenience in writing we shall henceforward omit the dashes in K, L, M and the suffix 't' in tensors.)

To obtain the reciprocal of $U - \beta\sigma$ we put

$$\left\{ U + \gamma L \left[U \vec{w}_L \right] - \gamma M \vec{w}_L \cdot \vec{w}_L \right\}^{-1} = \\ \left\{ AU + B \left[U \vec{w}_L \right] + C \vec{w}_L \cdot \vec{w}_L \right\} \quad . \quad (29)$$

Multiplying both sides by $U + \gamma L \left[U \vec{w}_L \right] - \gamma M \vec{w}_L \cdot \vec{w}_L$ and simplifying with the help of the relations

$$(\vec{w}_L \cdot \vec{w}_L) (\vec{w}_L \cdot \vec{w}_L) = w_L^2 (\vec{w}_L \cdot \vec{w}_L)$$

and

$$(\vec{w}_L \cdot \vec{w}_L) \cdot \left[U \vec{w}_L \right] = 0$$

we obtain

$$AU + B \left[U \vec{w}_L \right] + C \vec{w}_L \cdot \vec{w}_L + \gamma LA \left[U \vec{w}_L \right] + \\ \gamma LB \left\{ \vec{w}_L \cdot \vec{w}_L - w_L^2 U \right\} - \gamma MA \vec{w}_L \cdot \vec{w}_L - \gamma MC w_L^2 \vec{w}_L \cdot \vec{w}_L = U$$

Or equating the coefficients of U, $\left[U \vec{w}_L \right]$ and $\vec{w}_L \cdot \vec{w}_L$ we have

$$A - \gamma LB w_L^2 = 1 \\ \gamma LA + B = 0 \\ -\gamma MA + \gamma LB - \gamma MC w_L^2 + C = 0$$

which on solving gives

$$A = \frac{1}{1 + \gamma^2 L^2 w_L^2}, \quad B = -\frac{\gamma L}{1 + \gamma^2 L^2 w_L^2}$$

and

$$C = \frac{\gamma^2 L^2 + \gamma M}{(1 - \gamma M w_L^2)(1 + \gamma^2 L^2 w_L^2)}$$

whence we find from (29)

$$\begin{aligned} & \left\{ U + \gamma L [U \vec{w}_L] - \gamma M \vec{w}_L \cdot \vec{w}_L \right\}^{-1} \\ &= \frac{1}{1 + \gamma^2 L^2 w_L^2} \left\{ U - \gamma L [U \vec{w}_L] + \frac{\gamma^2 L^2 + \gamma M \vec{w}_L \cdot \vec{w}_L}{1 - \gamma M w_L^2} \right\} \end{aligned} \quad (30)$$

and therefore

$$\begin{aligned} (U - \beta \sigma)^{-1} &= \frac{1}{(1 - \beta K)(1 + \gamma^2 L^2 w_L^2)} \left\{ U - \gamma L [U \vec{w}_L] \right. \\ &\quad \left. + \frac{\gamma^2 L^2 + \gamma M \vec{w}_L \cdot \vec{w}_L}{1 - \gamma M w_L^2} \right\}. \end{aligned} \quad (31)$$

We therefore finally obtain

$$\begin{aligned} \epsilon' &= U + \beta^{-1} \{ (U - \beta \sigma)^{-1} - U \} \\ &= U + \frac{\gamma K}{\beta(1 + \gamma^2 L^2 w_L^2)} \left\{ \left(1 - \frac{\gamma L^2}{K} w_L^2 \right) U - \frac{L [U \vec{w}_L]}{K(1 - \beta K)} \right. \\ &\quad \left. + \frac{(\gamma L^2 + M) \vec{w}_L \cdot \vec{w}_L}{K(1 - \beta K)(1 - \gamma M w_L^2)} \right\}. \end{aligned} \quad (32)$$

or

$$\epsilon' = U + XYK \left\{ U - \frac{L [U \vec{w}_L]}{K(1 - \beta K)Y} + \frac{Z \vec{w}_L \cdot \vec{w}_L}{K(1 - \beta K)Y} \right\}. \quad (33)$$

where

$$X = \frac{1 - \beta K}{(1 - \beta K)^2 + \beta^2 L^2 w_L^2}, \quad Y = \frac{1 - \beta K - \frac{\beta L^2}{K} w_L^2}{1 - \beta K} \quad (34)$$

and

$$Z = \frac{\beta L^2 + M(1 - \beta K)}{1 - \beta K - \beta M w_L^2}$$

To find the equation for the refractive index, for a given direction of wave number in the medium, we shall consider

for simplicity the normal incidence only, and therefore put $\vec{n} = (0, 0, \mu)$. With the help of (14) and (33) the determinant of the components of $\epsilon^t - n^2 U + \vec{n} \vec{n}$ for the evaluation of μ^2 are found to be

$$\begin{vmatrix} 1 - \mu^2 + K_0 XY \left\{ 1 + \frac{w_z^2}{K_0(1 - \beta K)} \frac{Z}{Y} \right\} & \frac{L_0 X w_z}{1 - \beta K} & \frac{XZ}{1 - \beta K} w_z w_x \\ - \frac{L_0 X w_z}{1 - \beta K} & 1 - \mu^2 + K_0 XY & \frac{L_0 X w_z}{1 - \beta K} \\ XZ w_z w_x & - L_0 X w_x & 1 + K_0 XY \left\{ 1 + \frac{w_z^2}{K_0(1 - \beta K)} \frac{Z}{Y} \right\} \end{vmatrix} = 0 \quad (35)$$

It is to be remarked that only the diagonal elements of the integral tensors K , L and M appear in the determinant, and since the diagonal elements are equal we have denoted them simply by K_0 , L_0 and M_0 . They are given by

$$K_0 = \frac{4\pi}{i\omega} \frac{1}{3} K^t, \quad L_0 = \frac{4\pi}{i\omega} \frac{1}{3} L^t, \quad M_0 = \frac{4\pi}{i\omega} \frac{1}{3} M^t. \quad (36)$$

§ 2. To evaluate the expression for the refractive index we require to know the values of the integrals K_0 , L_0 and M_0 given by (36) and (11), (12) and (13). But unfortunately the general evaluation of these integrals is not possible. We shall, however, be content in the present investigation with the following simplest case.

(i) We do not make any hypothesis about the nature of the particles and therefore assume nothing about the mechanism of collisions between electrons and particles. We simply suppose Δ , the frequency of collisions, averaged somehow and take it to be constant throughout the range of integration.

(ii) We assume that the electron gas obeys non-degenerate statistics of Maxwell's, the distribution function f_0 being given by

$$f_0 = A e^{-e/kT} \quad . \quad . \quad . \quad (37)$$

This is evidently fulfilled in ionosphere where the electron concentration is very low.

Carrying out the integration we obtain

$$K_0 = -x_0 \frac{1 - \frac{i\Delta}{w}}{\left(1 - \frac{i\Delta}{w}\right)^2 - y^2}, \quad L_0 = -\frac{iK_0}{w} \frac{1}{1 - \frac{i\Delta}{w}} \quad (38)$$

and

$$M_0 = -\frac{K_0}{w^2} \frac{1}{\left(1 - \frac{i\Delta}{w}\right)^2}$$

where

$$x_0 = \frac{4\pi ne^2}{mw^2} \quad (39)$$

n being the electronic concentration.

Substituting these values in X, Y and Z and solving the determinant (35), we obtain after some deductions

$$1 - (\mu - ix)^2 = \frac{-\xi}{1 + \xi(1 - \tau_x^2)} \left[1 + \xi(1 - \tau^2) - \frac{1}{2}\tau_x^2 \pm \sqrt{\frac{1}{4}\tau_x^2 + \tau_x^2\{1 + \xi(1 - \tau^2)\}^2} \right] \quad (40)$$

where

$$\xi = \frac{-x_0 \left(1 - \frac{i\Delta}{w} + \beta x_0\right)}{\left(1 - \frac{i\Delta}{w} + \beta x_0\right)^2 - y^2}, \quad \tau = \frac{-y}{1 - \frac{i\Delta}{w} + \beta x_0} \quad (41)$$

$$\tau_x = \frac{-y_r}{1 - \frac{i\Delta}{w} + \beta x_0}, \quad \tau_z = \frac{-y_L}{1 - \frac{i\Delta}{w} + \beta x_0} \quad (42)$$

$$\xi(1 - \tau^2) = \frac{-x_0}{1 - \frac{i\Delta}{w} + \beta x_0} \quad (43)$$

and

$$y_L = \frac{eH_L}{mcw}; \quad y_r = \frac{eH_r}{mcw}; \quad y^2 = y_L^2 + y_r^2 \quad (44)$$

§ 3. One can easily verify that the equation (40) reduces to that of equation (67) in MI when we put $\beta = 0$. The

equation (40) agrees exactly with that given by Hartree and Appleton. The equation shows that for $\beta x_0 < 1$, *i.e.* for very short waves the polarisation term β can be neglected, whereas it becomes very important for $\beta x_0 > 1$, *i.e.* for very long waves. A little calculation will show that the behaviour of the refractive index with the incident frequency is altogether different in the latter case from that when $\beta = 0$; the range of low frequencies for which waves of one type of polarisation are propagated if $\beta = 0$ disappears if $\beta = \frac{1}{2}$. This has been already discussed and illustrated graphically by Hartree. It is thus from an investigation of the nature of the refractive index in the range of very long waves that we could decide the controversial point whether the polarisation correction should be included in investigating theoretical formulae for the refractive index in ionosphere. But unfortunately the experiments carried out in this region are very meagre and cannot at present give any definite solution to this controversy. Let us, however, hope that the experiments conducted with very long waves will in future throw much light on this controversial point.

It is to be remarked in conclusion that in the present investigation we find an expression for the refractive index, assuming that Δ , the frequency of collisions, is constant, *i.e.* it is independent on the energy of the incident electron. But, in fact, even in the simplest case of 'ideal ionosphere,' as considered in MI, which consists of electrons and ionised atoms only, the frequency of collisions Δ depends on the energy of the incident electron in a very complicated manner. The extension of the present investigation to the 'ideal ionosphere' will be undertaken in a subsequent paper.

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REFERENCES

- ¹ R. C. MAJUMDAR : *Zs. f. phys.* This paper will be referred to as MI.
- ² E. V. APPLETON : U.R.S.I. Reports (Washington, 1927), *Journ. Inst. Elec. Eng.*, vol. 71 (1932), p. 642.
- ³ D. R. HARTREE : *Proc. Camb. Phil. Soc.*, vol. 27 (1931), p. 143.
- ⁴ S. GOLDSTEIN : *Proc. Roy. Soc., A*, vol. 121 (1928), p. 260.
- ⁵ MARY TAYLER : *Proc. Phys. Soc.*, vol. 45 (1933), p. 245 ; *ibid.*, vol. 46 (1934), p. 408.
- V. A. BAILEY : *Phil. Mag.*, vol. 18 (1934), p. 516.
- D. F. MARTYN : *Phil. Mag.*, vol. 19 (1935), p. 376.
- H. G. BOOKER : *Proc. Roy. Soc., A*, vol. 147 (1934), p. 352 ; *ibid.*, vol. 150 (1935), p. 267.
- ⁶ L. TONKS : *Nature*, vol. 132 (1933), p. 101 ; *ibid.*, vol. 132 (1933), p. 710.
- ⁷ K. A. NORTON : *Nature*, vol. 132 (1933), p. 676.
- ⁸ D. R. HARTREE : *Nature*, vol. 132 (1933), p. 929.
- ⁹ C. G. DARWIN : *Nature*, vol. 133 (1934), p. 62 ; *Proc. Roy. Soc., A*, vol. 146 (1934), p. 17.

APPENDIX

f_0 = Fermi distribution function of the electron.

$$= \frac{1}{\frac{1}{A} e^{e/h\tau} + 1}$$

ϵ = energy of the electron.

k = Boltzmann's constant.

A = Sommerfeld's criterion for degeneracy

$$= \frac{nh^3}{2 (2\pi mkT)^{3/2}}$$

n = electron's concentration.

h = Planck's constant.

m = mass of the electron.

e = charge of the electron.

\vec{v} = group velocity = $\frac{\hbar}{m} \vec{K}$.

\vec{K} = wave vector of the electron.

\vec{F} = Lorentz force = $e\vec{E} + \frac{e}{c} [\vec{v} \vec{H}]$.

\vec{E} = intensity of the electric field = $\vec{E}_0 e^{i\omega t}$.

$\frac{\omega}{2\pi}$ = frequency of the wave.

grad \vec{K} = differentiation with respect to wave number \vec{K} .

τ = time of relaxation.

Δ = frequency of collisions.

μ = refractive index.

κ = extinction coefficient.

c = velocity of light.

H = resultant intensity of imposed magnetic field.

$\vec{w}_L = \frac{e\vec{H}}{mc}$ = double the Larmor frequency.

$y_L = \frac{eH_L}{mcw}$; $y_T = \frac{eH_T}{mcw}$; $y^2 = y_L^2 + y_T^2$.

H_L = components of external magnetic field along the direction of propagation.

H_T = components of external magnetic field transverse to the direction of propagation.

$\left(\frac{4\pi ne^2}{m}\right)^{\frac{1}{2}}$ = characteristic frequency of the medium.

$x_0 = \frac{4\pi ne^2}{mw^2}$.

